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# A Comparative Study on the Ecdysteroid Titre in the Normal, Decapitated and Thorax-ligated Larvae of Maize Stem Borer, *Chilo partellus* and Rice Moth, *Corcyra cephalonica* (Lepidoptera : Pyralidae)

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**Abstract:** The studies on the variations in circulating ecdysteroids in maize stem borer, *Chilo partellus* and rice moth, *Corcyra cephalonica* from the beginning of penultimate (fourth) larval stage until prepupal stage showed a major peak of hormone prior to ecdysis. Decapitation and thoracic-ligation significantly lowered the titre of circulatory ecdysteroids in both early-larval and late-larval instar larvae of *Chilo partellus* as well as *Corcyra cephalonica*.

**Keywords:** *Chilo*, *Corcyra*, decapitation, thoracic-ligation, ecdysteroids, radioimmunoassay

## INTRODUCTION

Characteristic changes occur in the titres of ecdysteroid during the postembryonic development of holometabolous insects including various lepidopterans (Smith, 1985; Steel and Vafopaulav, 1989; Kelly *et al.*, 1992). In the past decade, the maize stem borer, *Chilo partellus* and rice moth, *Corcyra cephalonica* (both of them belonging to the family pyralidae) have become most extensively investigated insects in India and other countries because of the severity of the damage that can result from these pests, which causes heavy loss of crop and stored products (Freeman, 1976; Esua, 1985; Chandurwar, 1989; Jayaraj *et al.*, 1994).

Studies in our laboratory mainly concentrate, on various endocrine factors which regulate/ influence different physiological processes during larval development and pupal-adult metamorphosis. Since, there is little titre information on the insects belonging to the family pyralidae, ecdysteroid titres during the larval development of *Chilo partellus* and *Corcyra cephalonica* were analysed. The present study also details the effect of surgical operations like decapitation and thoracic-ligation upon the titre of the ecdysteroids during the final instar larval development.

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\*Corresponding author

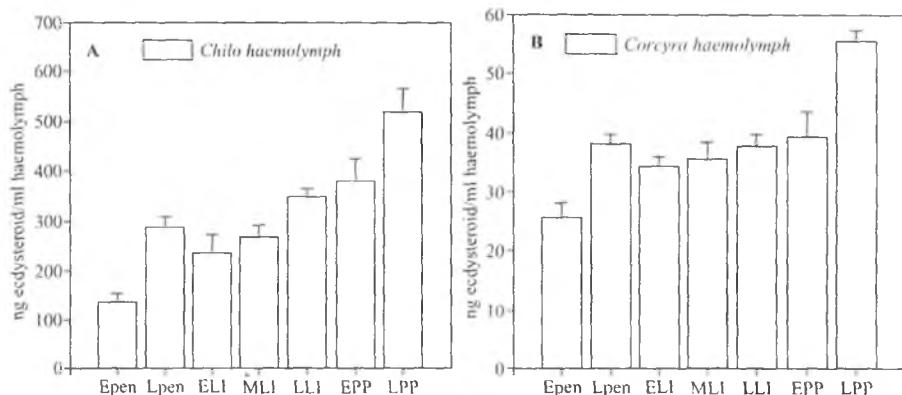


Fig. 1: Ecdysteroid titre during the larval development of *Chilo partellus* (A) and *Corcyra cephalonica* (B). Stages represented are early-penultimate (Epen), late-penultimate (Lpen), early-last instar (ELI), mid-last instar (MLI), late-last instar (LLI), early-prepupa (EPP) and late-prepupa (LPP). Each point represents mean  $\pm$  SD of three separate determinations of duplicate aliquots.

## MATERIAL AND METHODS

### Insects

*Chilo partellus* larvae were reared on a semisynthetic diet (Taneja and Leuschner, 1985) in a culture room at  $26 \pm 1^\circ\text{C}$  temperature, 14 : 10 h LD period and  $70 \pm 5\%$  relative humidity. The culture of *Corcyra cephalonica* was maintained on coarsely crushed sorghum seeds under the above mentioned conditions. The larvae of *Chilo partellus* as well as *Corcyra cephalonica* were staged as previously described (Alghali, 1985; Ashok and Dutta-Gupta, 1988; Ismail and Dutta-Gupta, 1988). For decapitation and thoracic-ligation experiments, early-last and late-last instar of *Chilo partellus* and *Corcyra cephalonica* were used.

### Radioimmunoassay

Ecdysteroids were extracted from the larval haemolymph collected by cutting the prolegs and diluted with two volumes of ice cold methanol as described previously (Bollenbacher *et al.*, 1981). The antiserum (2-B) a gift from Dr. W. E. Bollenbacher [University of North Carolina, U.S.A] was used for radioimmunoassay (RIA). Microassays were performed according to the procedure of Warren *et al.* (1984) with slight modification. The RIA data is expressed in 20 hydroxyecdysone (20E) equivalents since this ecdysteroid was used as the standard. The labelled ligand for the RIA was [23-24-<sup>3</sup>H] ecdysone [60 Ci/mmol], and was obtained from New England Nuclear.

## RESULTS

The present study showed the presence of a low titre of ecdysteroids in the early-penultimate instars (early-fourth larval) of *Chilo partellus* as well as *Corcyra cephalonica* which increased in the late-penultimate instar. There was a slight decline in the

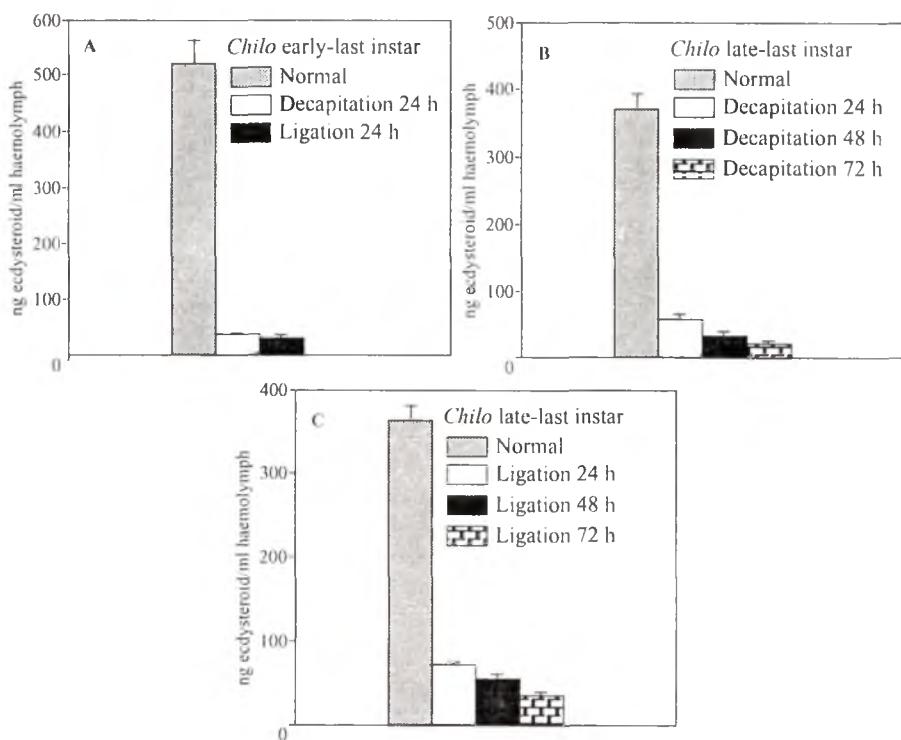


Fig. 2: Effect of decapitation and thoracic-ligation on ecdisysteroid titre after 24, 48 and 72 h in early (A) and late-last instar (B, C) larvae of *Chilo partellus*. Each determination represents mean  $\pm$  SD of three separate determinations of duplicate aliquots.

early-last larval instar but it increased once again gradually through mid-last, late-last instar larval and early and late-prepupal stages of development. The end of the larval development was marked by a high hormonal titre (Fig. 1A, B).

Decapitation and thoracic-ligation experiments in early (Fig. 2A) and late-last instar larvae (Fig. 2B, C) of *Chilo partellus* revealed that the titre of ecdisysteroids declined significantly after 24 h. Furthermore, this decline was more pronounced at longer periods of 48 and 72 h post-ligation (Fig. 2B, C). In *Corcyra cephalonica* also, decapitation (Fig. 3A, C) and thoracic-ligation (Fig. 3B, D) caused a decline in the titre of ecdisysteroids in early as well as late-last instar larval stages. Once again, the decline was more pronounced with longer periods at 48 and 72 h post-ligation. However, in this case the decline was gradual and even after 72 h a sufficient amount of ecdisysteroids remained, in the haemolymph.

## DISCUSSION

*Chilo partellus* as well as *Corcyra cephalonica* have a classical ecdisysteroid pattern i.e., a major peak during the later part of each stage, just preceding each ecdysis

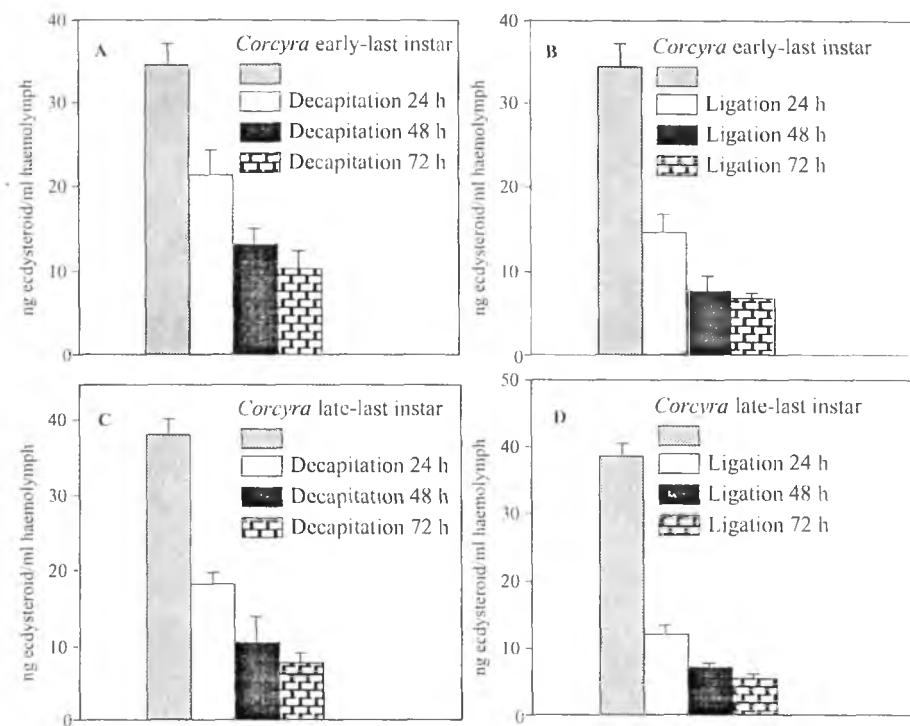


Fig. 3: Effect of decapitation and thoracic-ligation on ecdysteroid titre after 24, 48, and 72 h in early (A, B) and late-last instar (C, D) larvae of *Corcyra cephalonica*. Each determination represents mean  $\pm$  SD of three separate determinations of duplicate aliquots.

as demonstrated earlier by several workers (Bollenbacher *et al.*, 1981; Planteron *et al.*, 1984; Smith, 1985; Newitt and Hammock, 1986; Gelman *et al.*, 1988; Yang *et al.*, 1995). The increase during the last instar larval development was more pronounced than in the penultimate instar. The quantitative changes observed in ecdysteroid, regulate molting and metamorphosis through a cascade of gene expression and repression events. It is interesting to note that the total concentration of ecdysteroids in the haemolymph of *Chilo partellus* was nearly 8–10 fold high at all the developmental stages when compared with *Corcyra cephalonica*. Significant variation (2 to 20 fold) in haemolymph ecdysteroid titre has already been well reported in Lepidopteran insects (Dean *et al.*, 1980).

Prothoracicotropic hormone (PTTH) secreted by brain plays an important role in the regulation of steroidogenesis in insects (Bollenbacher and Granger, 1985). In the present study decapitation and thoracic-ligation caused a significant decline in the haemolymph ecdysteroid titre of both *Chilo partellus* and *Corcyra cephalonica*. Decapitation isolates the prothoracic gland from PTTH influence leading to reduced synthesis of ecdysone (Dean *et al.*, 1980). On the other hand, thoracic-ligation eliminates total prothoracic gland which is the primary source of ecdysone in insects. The decline

was slow and gradual in *Corcyra cephalonica*, while it was sharp in *Chilo partellus*. The observed difference in the pattern might be due to the short half life of circulating ecdysteroids in *Chilo partellus* when compared with *Corcyra cephalonica* (Koolman, 1982, 1990; Rees, 1995). In addition, there might be some other organs/tissues localised in the posterior part of the larval body of the *Corcyra cephalonica* which might accumulate and release and/or synthesise and release these ecdysteroids in the haemolymph of decapitated and thorax-ligated insects. Studies in Lepidoptera reveal that besides, prothoracic glands, several other tissues like testis, epidermis, midgut and oenocytes could be alternative sites of ecdysteroid production during postembryonic development (Loeb *et al.*, 1988; Delbecque *et al.*, 1990; Jarvis *et al.*, 1994). However, detailed investigations are required to substantiate this proposition. Sakurai *et al.* (1991) have reported the presence of high titre of haemolymph ecdysteroids in the isolated pupal abdomen of *Maclura sexta* which were devoid of testis as well as digestive tract. The mechanisms involved in controlling moulting in isolated abdomens as well as the sources of relatively high titres of ecdysteroids in these abdomens are open areas of investigation. The possible significance of ecdysteroid titres in the regulation of various physiological processes during the postembryonic development of *Chilo partellus* and *Corcyra cephalonica* will be the subject of future research in our laboratory.

#### ACKNOWLEDGEMENTS

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# Cuticular Hydrocarbons and Biosystematics of Entimine Weevil Genera *Lepropus* Schoenherr and *Brachyaspistes* Fahraeus (Curculionidae: Coleoptera)

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**Abstract:** Critical studies were undertaken to evaluate the conflicting views on the biosystematics of the entimine weevil genus *Lepropus* Schoenherr, especially regarding its relationship with other entimine generic taxa namely *Brachyaspistes* Fahraeus, *Lepidospyris* Marshall and *Indomias* Marshall. These include analyses of morphotaxonomic evidences, zoogeographical distribution and molecular systematic studies on the cuticular hydrocarbons. These analyses have concluded that *Lepropus* and *Brachyaspistes* are distinct genera; *Lepidospyris* and *Indomias* overlap with *Lepropus* and their status needs reconsideration; and molecular systematic studies involving cuticular hydrocarbons have a significant role to play in species diagnosis.

**Keywords:** Biosystematics, *Lepropus*, *Brachyaspistes*, Curculionidae, Coleoptera, Cuticular hydrocarbons

## INTRODUCTION

The entimine weevil genus *Lepropus* was erected by Schoenherr (1826) with *lateralis* (Fabricius, 1792) as the type. Faust while reclassifying this described three genera namely *Lepidastycus* (1895), *Astycophilus* and *Astycophobus* (1897). Marshall (1916) synonymized *Brachyaspistes* Fahraeus (in Schoenherr 1840) with this and redefined the same. He endorsed Heller (1908) and opined that Faust's genera were based on characters subject to serious exceptions. Emden (1936) divided it into five subgenera namely *Astycomerus* Kolbe (1883), *Heteroscapus* Faust (1886), *Ischnotrachelus* Schoenherr (1840), *Brachyaspistes* and *Astycus* s.str. Emden and Emden (1939) added another subgenus *Ischnotracheloides* Hustache. Emden (1944) followed this but dropped the subgenus *Ischnotracheloides*. Many anomalies resulted due to such revisions which disregarded some major taxonomic characters and few of these were

pointed out by Marshall (1916). Other eminent weevil systematists like Voss (1962) and Hoffmann (1966, 1968) treated *Ischnotrachelus* a distinct genus, resulting in conflicting views on Emden's revision.

Taking into account these conflicting views and as a result of the recent revision (Poorani and Ramamurthy, 1997), there are at present 36 species of *Lepropus* known from the Oriental region. Of these 18 each fall under the subgenera *Lepropus* s.str., and *Brachyaspistes*. The main objective of the present study is to investigate these conflicting views on the biosystematics of *Lepropus* by analyzing the morphological and zoogeographical evidences and supplement the same with cuticular hydrocarbon analyses. Incidentally the utility of cuticular hydrocarbons in species diagnosis is also evaluated and results discussed.

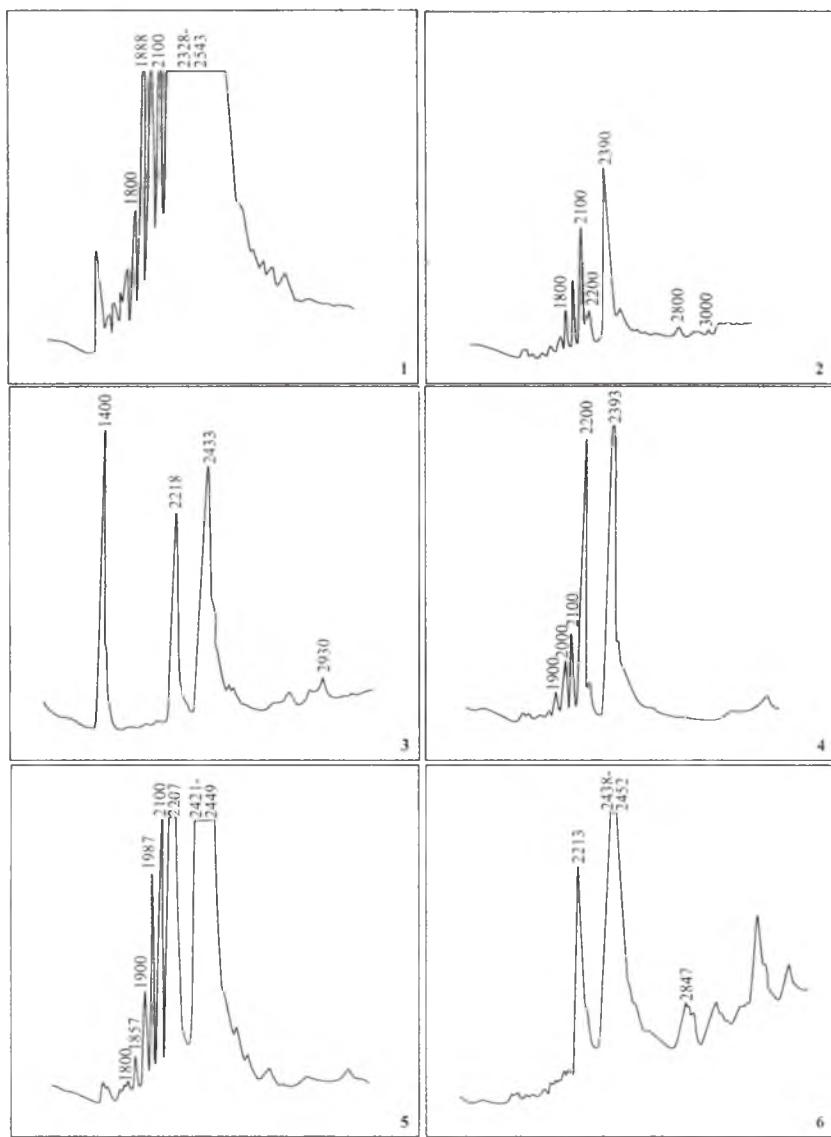
#### MATERIAL AND METHODS

To analyse the morphological variations the taxonomic characters were studied taking into account the methods envisaged in Poorani and Ramamurthy (1997). For the analysis of cuticular hydrocarbon profile, the cuticular wax was extracted by soaking the cuticle of individual insects with 2 ml of freshly distilled n-hexane for 24h. The extracts were collected by decantation followed by rinsing with additional 2 ml hexane and were concentrated *in vacuo* to remove hexane. The weights of the leftover residue (wax) were recorded. The cuticular wax was dissolved exactly in 1 ml hexane. A 10 microlitre solution was used for hydrocarbon analysis by Hewlett Packard model 5890 Series II gas liquid chromatograph fitted with a megabore column (10m × 0.53 mm id) packed with OV-17 and equipped with flame ionization detector (FID) and a Hewlett Packard model 3380A integrator. The column temperature regime was programmed as follows; initial temperature maintained at 50°C for 1.75 min, subsequent rate of increase @ 25°C/min upto 180°C for 1 min, maintained isothermal at 180°C for 1 min and heated further @ 8°C/min to 270°C. Kovats' retention indices (KI) were assigned to the sample peaks using authentic hydrocarbon standards (C<sub>5</sub>- C<sub>30</sub> n-alkanes procured from Aldrich, USA). Detector and injector temperatures were maintained at 250°C respectively. Nitrogen was used as carrier gas at a flow rate of 20 ml/min. A peak that co-eluted with n-pentacosane (n- C<sub>25</sub>H<sub>52</sub>) was assigned KI value of 2500. The KI values were computed following the method of Kovats (1965) and the cuticular waxes from different species were compared.

#### RESULTS AND DISCUSSION

##### Morphotaxonomic evidences

Further to the revision of the *Lepropus* from the Oriental region (Poorani and Ramamurthy, 1997), additional morphological characters were investigated. These have led to several additional differentiating characters between *Lepropus* s.str., and *Brachyaspistes*. In addition to the characters like number of setae on mentum and mandibular scrobes which are usually employed in differentiating genera of this subfamily, several additional morphological characters were identified as differentiating them. These characters as summarized in Table 1, indicate that the dissimilarities overplay similarities and it is imminent to consider these as distinct genera. Several taxonomic characters provided by scrobes, carination of rostrum, sinuation of posterior margin

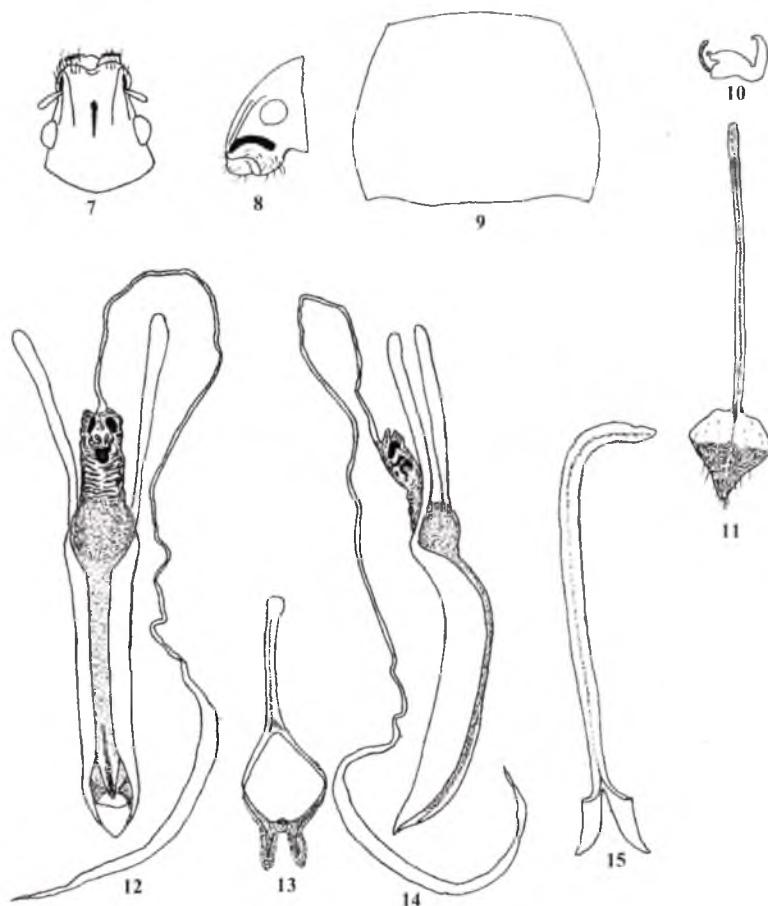


Figs. 1-6. *Lepropus*. Gas chromatograms of cuticular extracts of 1: *chrysoclorus*; 2: *christopheri*; 3: *siamensis*; 4: *lateralis*; 5: *lateralis* var. *rutilans*; 6: *aurovittatus*.

of prothorax, setae on mentum, genitalia and hemisternites of ovipositor substantiate their distinction as genera to make them monophyletic and their classification natural.

#### *Lepropus vis-a-vis other closely allied entimine genera*

Two entimine genera namely *Lepidospyris* Marshall and *Indomias* Marshall resemble the genus *Lepropus*. The members of the former apparently resemble the smaller grey



Figs. 7-15 *Lepidospyris demissa* Marshall. 7: Head, dorsal view; 8: Head, lateral view; 9: Prothorax, dorsal view; 10: Spermatheca; 11: Spiculum ventrale; 12: Aedeagus, dorsal view; 13: Tegmen; 14: Aedeagus, lateral view; 15: Spiculum gastrale.

coloured species of *Lepropus* but can be distinguished by the squamose apices of hind tibiae, embossed scales on the dorsal surface and the scrobes curving downwards in front of the eyes in addition to genitalia (Figs. 7-15). Many species of *Lepropus* possess characters such as widely separated forecoxae, metasternum longer than mesocoxae and tibiae without dorsal furrow, embossed scales [*lewisi* Marshall (1916)], tibiae with squamose tarsal grooves [*chrysoclorus* (Wiedemann, 1823), *janakiramani* Poorani and Ramamurthy (1997), *venkataramani* Poorani and Ramamurthy (1997), *thompsoni* Poorani and Ramamurthy (1997), *siamensis* Poorani and Ramamurthy (1997) and *swarajae* Poorani and Ramamurthy (1997)] and female genitalia (all species falling under *Brachyaspistes*) which overlap with those of *Lepidospyris*. The other look alike genus *Indomias* has characters such as the presence of two setae on the mentum, sec-

Table 1: *Lepropus* s.str., vis-a-vis *Brachyaspistes*

Similarities	Character	Dissimilarities			<i>Brachyaspistes</i>
		<i>Lepropus</i> s. str.			
Elytra with well developed shoulders, sinuate basal margin, lateral notch to receive the head of metasternal episternum	No. of setae on mentum	Always more than four			Mostly two and rarely four
Mesosternal epimeron and episternum subequal	Prothorax	Median present	furrow	never	Always with a median furrow
		Posterior thicker	margin	variable,	Always bisinuate, fine
	Tibiae	With or without a dorsal furrow or sulcus			Never sulcate
	Tibial apex	An outer bevel with two rows of setae and an inner flange present			Only outer bevel with two rows of setae present
	Elytral vestiture	Flat scales always ridged			Flat scales never ridged
	Female genitalia	Spermatheca subglobose, spiculum ventrale 1.5–2.5x as long as basal plate			Spermatheca subcylindrical, spiculum ventrale 3–5x as long as basal plate
	Distribution	South and Southeast Asia (except Sri Lanka)			Peninsular India and Sri Lanka
	Cuticular Hydrocarbon profile	C <sub>14</sub> –C <sub>20</sub> hydrocarbons present in more quantities			C <sub>14</sub> –C <sub>20</sub> hydrocarbons only in traces

ond ventrite longer than the third and fourth together, distinct scutellum and the structure of female genitalia, especially spiculum ventrale overlapping, particularly with the members of *Brachyaspistes*. All these conclude that there is a need to reconsider the status of *Lepidospyris* and *Indomias*.

### Zoogeographical distribution

The geographical boundaries of *Lepropus* and *Brachyaspistes* are well defined with the former distributed in South and South East Asia (except Sri Lanka) and the latter confined to Sri Lanka and peninsular India. In India *Lepropus* s.str., is found mainly in the north and the north eastern regions and only *oculatus* (Heller, 1908) and *lateralis* had been recorded from the south so far (Table 1). In the present study we came across a specimen from New Guinea of *L. (L.) adamsoni* (Marshall, 1916), a new record for the Australian region. The inclusion of *angustulus* Fairmaire (1888), an African species in this subgenus remains questionable and needs review.

### Cuticular hydrocarbon studies

#### *Lepropus vis-a-vis Brachyaspistes*

The cuticular hydrocarbon profile of *Lepropus* s.str., and *Brachyaspistes* obtained by analysing hexane extracts of whole insects by GLC were found to be distinct (Table 1). In *Lepropus* s.str., C<sub>14</sub>-C<sub>20</sub> compounds are present in significant quantities (Figs. 1-5), whereas in *Brachyaspistes*, these are found only in traces (Fig. 6). All the species analysed namely *L. (L.) chrysoclorus*, *L. (L.) christopheri* Poorani and Ramamurthy (1997), *L. (L.) siamensis*, *L. (L.) lateralis* and *L. (B.) aurovittatus* (Heller, 1908) showed unique profiles (Figs. 1-6). All the species of *Lepropus* s.str., were found to have profiles ranging from KI 1400-2200, whereas in *Brachyaspistes* these hydrocarbons were only in traces and those with KI 2200-2900 were in large quantities.

#### *Species diagnosis*

In general cuticular hydrocarbons having C<sub>14</sub> to C<sub>30</sub> carbons were detected in the species tested. The species namely *L. christopheri* and *L. chrysoclorus* which were morphologically similar and difficult to distinguish even by genitalic characters (Poorani and Ramamurthy, 1997) were compared for their cuticular hydrocarbons. The profile of *chrysoclorus* (Fig. 1) showed a very large dominant peak consisting of several undifferentiated peaks at KI 2328-2543, which were conspicuously absent in the profile of *christopheri* (Fig. 2). Peaks with KI 1800 and 2100 which coeluted with n-octadecane (C<sub>18</sub>) and n-heineicosane (C<sub>21</sub>) are potential species markers as these were quantitatively quite different. The compound with KI 1800 was present to the extent of approximately 0.68% and 0.08% of the body weight of *chrysoclorus* and *christopheri* respectively, while the compound with KI 2100 comprised 0.20% and 0.02% respectively. The profile of *christopheri* was characterized by two more peaks with KI 2200 and 2800, coeluting with n-docosane (C<sub>22</sub>) and n-octacosane (C<sub>28</sub>) respectively, which were absent in *chrysoclorus*. Repeat analyses showed slightly different profiles which were again distinctly different for both the species. In case of *siamensis* the profile showed three major peaks with KI 1400 [coeluting with n-tetradecane (C<sub>14</sub>)], 2218 and 2433 and a smaller peak with KI 2930 and was distinct from those of the rest. Similarly chromatograms of *aurovittatus* revealed three major peaks with KI 2213, 2438-2452 and 2847. These results are promising and definite identification of these compounds requires tandem technique of mass spectral analysis. Although repeat analyses in the same species sometimes revealed variations, by sampling a natural population of each species the effect of such variations can be minimized. From these studies, it can be concluded that successful identification of species by their cuticular hydrocarbon profiles is indeed possible.

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## Resistance of *Trogoderma granarium* Everts to Phosphine

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**Abstract:** Populations of *Trogoderma granarium* Everts collected from different locations were inbred in the laboratory. The larvae and adults from F<sub>1</sub> generations after exposure to graded concentrations of phosphine for 20 hr were kept under observation till the end-point mortality was attained. The LC<sub>50</sub> values of phosphine for larvae and adults showed variations in tolerance between different populations of *T. granarium*. The strain collected from the farmer's house, village Balala (BTG), Jalandhar was found to be the most susceptible whereas that from another farmer's house, Kotkapura (KTG), Faridkot was the most tolerant to phosphine. Adults of *T. granarium* of KTG were about 6.45 fold tolerant to phosphine in relation to BTG. The selections of third-instar larvae of KTG for three consecutive generations resulted in 2-fold increase in its tolerance compared to its parent population and about 7-fold to that of BTG. Adults of selected strain also showed a fairly high degree of resistance (×12–17) thereby suggesting that phosphine resistance in this species can be enhanced with relative ease.

**Keywords:** Phosphine, Resistance, *Trogoderma granarium*, Strain

### INTRODUCTION

The Khapra beetle, *Trogoderma granarium* Everts is a devastating pest of stored products, particularly cereals and seeds. This beetle has become widely distributed with increased trade and movement of food stuffs (Banks, 1977). It is particularly abundant in the Indian sub-continent, parts of North Africa and the Middle East. It also occurs in Central America and South East Asia as well as in heated storage premises in temperate regions of Europe and North America (Burges, 1959).

*Trogoderma granarium* has been found to be an extremely difficult pest to control because of natural tolerance of its larvae to insecticides (Pradhan and Bhatia, 1956; Gupta *et al.*, 1971; Judal, 1985). Fumigation with phosphine has been found to provide satisfactory control of this insect species (Vincent and Lindgren, 1972; Hole *et al.*, 1974; El-Lakwah, 1978), but it has also been known that some developmental stages of this species especially young eggs and diapausing larvae (Vincent and Lindgren, 1972; Hole *et al.*, 1974; Bell *et al.*, 1984) are quite tolerant of phosphine. Besides the natural tolerance of *T. granarium* to insecticides and phosphine, Borah and Chahal (1979) have

Table 1: Location and year of collection of different populations of *T. granarium*

Location (Source)	Year of collection	Symbol
Balala Village, Jalandhar Distt. (Farmer's house)	1980	BTG
Ludhiana City (Laxmi Flour Mill)	1980	LTG
Ludhiana City (Laxmi Flour Mill)	1984	LFTG
Dangon Village Ludhiana Distt. (Farmer's house)	1988	DTG
Jagraon Ludhiana Distt. (FCI Warehouse)	1988	JTG
Mallah Village Ludhiana Distt. (Farmer's house)	1988	MTG
Kotkapura Faridkot Distt. (Farmer's house)	1988	KTG

reported the development of resistance in various developmental stages of this species. However, Winks (1986a) was skeptical of this report, because of short post-treatment holding period and many errors in the calculation (Bell *et al.*, 1984). Studies were, therefore, undertaken to further investigate the distribution and frequency of phosphine resistance in *T. granarium* at sufficiently long post-treatment holding period.

## MATERIAL AND METHODS

### Insects

Insect cultures of the different populations/strains of *Trogoderma granarium* Everts collected from different locations in Punjab, India (Table 1) were maintained in separate glass jars (15 × 10 cm) on sterilized wheat grains (broken/whole) and 0.5% yeast extract at 37 ± 0.5°C and 70% relative humidity (Solomon, 1951). The larvae after removing excreta, exuviae and wheat grains were transferred at regular intervals of about 30 days to the fresh food. The larvae of each population/strain were separated from culture jars utilizing their thigmotropic response by spreading wheat grains and insect contents thinly on filter paper. After 2–3 minutes, the grains were transferred back to the jars and the larvae sticked to the filter papers were collected with Camel's hair brush. The overcrowding of larvae was avoided by releasing only 200–300 larvae to glass jars about half filled with food grains. The pupae of different populations/strains were separated and subsequently used for raising the single pair *F*<sub>1</sub> progenies.

### Preparation of insects for treatments

In order to obtain the larvae of same age group in a particular stage, about 1000 freshly emerged adults of each population/strain were released separately in glass jars (15 × 10 cm) having wire net at once end and muslin cloth at the other end. The jars were

placed over the Petri-dishes (150 × 20 mm) with the end having wire net resting over them. The eggs laid within 24 hr were collected in the Petri-dishes and allowed to hatch to first-instar larvae. These larvae were kept individually in glass vials (14 × 50.8 mm) containing broken wheat grains and yeast extract. Appropriate number of such vials were kept to maintain the continuous supply of the culture. Based on moulting, third- and fifth-instar larvae were collected. Pupae of same age group were obtained by releasing about 300–500 full grown larvae of each population/strain per jar provided with grains. The adults emerged within 24 hr were considered as fresh.

#### Bioassay method

Phosphine gas was prepared by hydrolysis of aluminium phosphide (Celphos<sup>R</sup>) tablets according to FAO method (Anonymous, 1975). Four replicates of insects of desired stage (i.e. third- and fifth-instar larvae and adults) in Petri-dishes (50 × 15 mm) were placed in fumigation chambers (2.5 l capacity) maintained at  $37 \pm 0.5^\circ\text{C}$  and 70% RH. Each replicate comprised of 20 insects. Batches of insects belonging to corresponding stage of each population/strain were also kept as control.

Graded concentrations (0.1–3.0 mg/l) of 86% phosphine mixture were injected into the air tight fumigation chambers using air-tight Hamilton Syringes (1.0 and 2.5 ml capacity). The quick circulation of gas in the fumigation chambers was ensured by slowly pumping of a 10 ml gas-tight syringe inserted in the chamber. After exposure for 20 hr the insects were removed from the fumigation chambers and were transferred to Petri-dishes (100 × 17 mm) containing broken/whole wheat grains and yeast extract. During the post-treatment holding period, the insects were kept under the identical conditions as mentioned above. Mortalities were recorded daily in case of adults till the end-point was reached whereas in case of larvae, the mortalities were recorded at 5-day intervals till the adults emerged from the survivors. The LC<sub>50</sub> values of phosphine for different developmental stages of each population/strain were derived by Probit analysis (Finney, 1971).

Population of KGT was further selected with phosphine by exposing third-instar larvae to 0.956 mg/l for 20 hr for three consecutive generations under laboratory conditions. Twenty batches of 50 insects each were exposed. The survivors after third generation were multiplied by using single pair. The resistance of third- and fifth-instar larvae and adults to phosphine of this selected strain was evaluated as mentioned above.

#### RESULTS

The LC<sub>50</sub> values of phosphine calculated from end-point mortality data for third- and fifth-instar larvae and adults showed variations in tolerance between populations/strains of *Trogoderma granarium* Everts collected from different locations in the Punjab (Table 2–4 and Fig. 1). The population collected from the farmer's house, village Balala (BTG), Jalandhar was the most susceptible (LC<sub>50</sub>: 0.219; 0.420 and 0.112 mg/l respectively for third- and fifth-instar larvae and adults), whereas that from another farmer's house, Kotkapura (KTG), Faridkot was the most tolerant to phosphine (respective LC<sub>50</sub>: 0.770, 2.188 and 0.723 mg/l). Resistance ratios for third-instar larvae of various populations/strain with respect to reference Balala (BTG) population, ranged from 2.32 to 3.51 (Table 2), whereas for fifth-instar larvae the range was from 1.65 to 5.21 (Table 3). The resistance ratio in case of adults of various populations ranged from 3.69 to 6.45

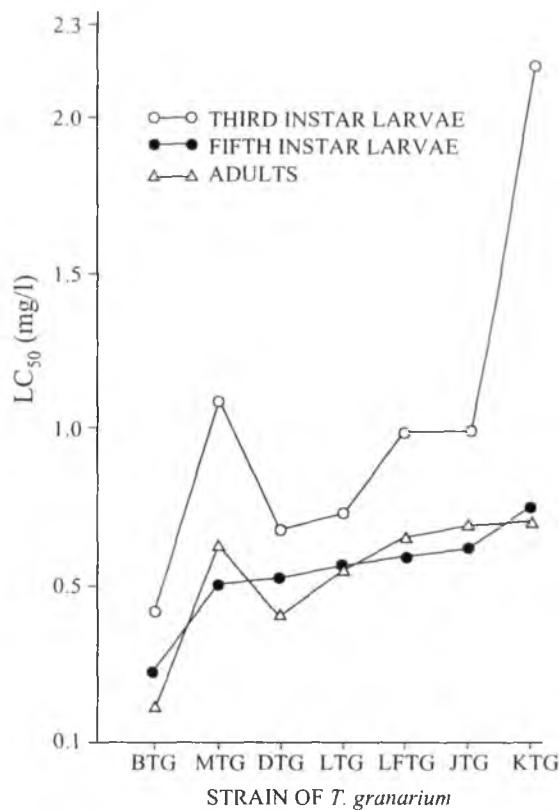


Fig. 1: Showing tolerance of different stages of different strains of *T. granarium* to phosphine

(Table 4). In general the populations/strains showed the same pattern of phosphine-tolerance in different developmental stages. However, the population of MTG did not show consistent tolerance to phosphine. The degree of resistance to phosphine in adults was more than in the immature stages.

The resistance ratio of the phosphine-selected KTG strain showed that adults (males and females) and immature stages (third- and fifth-instar larvae) manifested almost 2 times resistance of phosphine compared to its parent population (Table 5). The third- and fifth-instar larvae and adults stages of KTG had 7.41, 9.96 and 12.71 to 17.14 times respectively more resistance in relation to BTG strain.

## DISCUSSION

The degree of tolerance/resistance to phosphine manifested by different stages (2.32–6.45) of different populations/strains *T. granarium* is low (Table 2–4). In contrast, Borah and Chahal (1979) reported a 40-fold increase in tolerance for first-instar larvae of *T. granarium* collected from Jagraon Punjab. Bell *et al.* (1984) pointed out that many of LD<sub>50</sub> and LD<sub>90</sub> values reported by Borah and Chahal (1979) were wrong ac-

Table 2: Dosage estimates and parameters of regressions of probit mortality on log-concentration of phosphine for third-instar larvae of various strains of *T. granarium* collected from Punjab.

Strain	LC <sub>50</sub> (mg/l)	Fiducial limits (mg/l)		Slope $\pm$ S.E.	Heterogeneity		Resistance ratio
		Lower	Upper		df	$\chi^2$	
BTG	0.219	0.158	0.263	2.73 $\pm$ 0.51	2	0.50	1.00
MTG	0.508	0.331	0.602	2.48 $\pm$ 0.57	2	3.43	2.32
DTG	0.531	0.371	0.616	2.84 $\pm$ 0.58	2	1.62	2.42
LTG	0.576	0.407	0.708	3.17 $\pm$ 0.82	1	2.48	2.63
LFTG	0.602	0.407	0.708	3.18 $\pm$ 0.82	1	3.40	2.75
JTG	0.636	0.398	0.794	2.90 $\pm$ 0.92	1	1.29	2.90
KTG	0.770	0.490	0.933	2.09 $\pm$ 0.74	2	2.78	3.51

Table 3: Dosage estimates and parameters of regressions of probit mortality on log-concentration of phosphine for fifth-instar larvae of *T. granarium* collected from the Punjab

Strain	LC <sub>50</sub> (mg/l)	Fiducial limits (mg/l)		Slope $\pm$ S.E.	Heterogeneity		Resistance ratio
		Lower	Upper		df	$\chi^2$	
BTG	0.420	0.302	0.724	1.83 $\pm$ 0.50	1	1.78	1.00
DTG	0.682	0.489	1.071	2.13 $\pm$ 0.58	1	3.66	1.65
LTG	0.742	0.339	1.412	2.60 $\pm$ 0.56	2	0.78	1.76
LFTG	1.000	0.794	1.318	2.97 $\pm$ 0.93	1	2.30	2.38
JTG	1.006	0.851	1.412	3.36 $\pm$ 0.78	1	2.60	2.39
MTG	1.095	0.912	1.445	4.29 $\pm$ 1.04	1	2.73	2.60
KTG	2.188	1.905	2.399	7.44 $\pm$ 2.04	1	1.34	5.21

cording to their quoted regression line equations. For instance, a 40-fold resistance was infact less than  $\times 20$ . The maximum increase in tolerance in *T. granarium* during the present study was observed to be about 6-fold at end-point mortality and the post-treatment holding period in the toxicity of phosphine is an important factor due to the fact that mortalities of adults and larvae obtained at end-point were substantially high compared to short post-treatment holding period. The post-treatment holding period for adults and larvae varied from 5–8 and 17–26 days respectively. Borah and Chahal (1979) recorded the mortality after short post-treatment holding period which

Table 4: Dosage estimates and parameters of regressions of probit mortality on log concentration of phosphine for adults of *T. granarium* collected from the Punjab

Strain	LC <sub>50</sub> (mg/l)	Fiducial limits (mg/l)		Slope $\pm$ S.E.	Heterogeneity		Resistance ratio
		Lower	Upper		df	$\chi^2$	
BTG	0.112	0.079	0.174	2.42 $\pm$ 0.68	2	0.71	1.00
DTG	0.413	0.284	0.533	3.23 $\pm$ 1.18	1	1.85	3.69
LTG	0.561	0.473	0.668	5.18 $\pm$ 1.86	1	2.53	5.00
MTG	0.639	0.508	0.682	5.86 $\pm$ 1.72	1	0.66	5.70
LFTG	0.669	0.564	0.774	4.25 $\pm$ 1.66	1	3.32	5.97
JTG	0.706	0.586	0.816	3.90 $\pm$ 1.64	1	1.55	6.30
KTG	0.723	0.582	0.901	3.55 $\pm$ 1.25	2	2.51	6.45

Table 5: LC<sub>50</sub> and resistance ratio of phosphine selected strain (KTG) in relation to reference strain (BTG) and its parent population after exposure to 20 hour.

Stage	Strain	LC <sub>50</sub> (mg/l)	Resistance ratio with respect to	
			BTG	KTG parent
Adult (female)	BTG	0.137	1.00	—
	KTG selected	1.742	12.71	2.42
Adult (male)	BTG	0.080	1.00	—
	KTG selected	1.371	17.14	1.90
Adult (mixed)	KTG parent	0.719	—	1.00
	BTG	0.212	1.00	—
Third-instar larvae	KTG parent	0.778	3.66	1.00
	KTG selected	1.571	7.41	2.02
	BTG	0.458	1.00	—
Fifth-instar larvae	KTG parent	2.179	4.75	1.00
	KTG selected	4.562	9.96	2.09

did not take into consideration the speed of action of phosphine, hence, the degree of resistance measured was high. Further, the use of fixed post-treatment holding periods in laboratory tests designed to measure the acute response of insects to poison is untenable unless it is known that the time chosen is sufficient for all the individuals, when tolerance has been exceeded, to respond to the dose applied (Winks, 1986a). For instance, Beard (1949) investigated the effect of different times of assessment on measurement of the toxicity of arsenic and parathion injected into the body cavity of milkweed bug, *Oncopeltus fasciatus* (Dall). It was found, on the basis of observations made two days following treatment that arsenic was almost 250 times more toxic than parathion whereas at end-point the difference was less than 3-fold. Similarly, Winks (1986b) showed that when assessment made before end-point was reached, large differences were obtained in mortality levels and in parameters of probit lines fitted to the data. It was further found that if different concentrations were used for the susceptible and resistant strains, comparisons made before the end-point reached could be grossly misleading.

The selection of third-instar larvae of KTG population/strain for three consecutive generations with phosphine increased its resistance to 7-fold in relation to BTG strain and 2-times as resistant as its parent population. Thus, there is adequate evidence that phosphine-resistance in this species can be selected with relative ease, because of this, it may pose a real threat to the continued use of this fumigant for its control. The resistant ratio manifested at different development stages (adults and larvae) after selection was fairly high and the adequate management strategies may be adopted to reduce the selection pressure and prolong the effective life of this useful fumigant.

Similar observations were also made by other workers in case of other stored grain insect pests such as *Tribolium castaneum* Herbst, *Rhyzopertha dominica* (F.) and *Sitophilus granarius* (L.) etc. ((Winks, 1986a), Taylor, 1986; 1989, Taylor and Halliday, 1986; Zettler *et al.*, 1989). According to Winks (1986a) it is possible that phosphine resistance can be selected in most if not all, strains of all species of insects and the genetic material for resistance may be present in all strains of all species of insects and the genetic material for resistance may be present in all strains or that mutation frequency is higher than expected. The slope of log- concentration probit re-

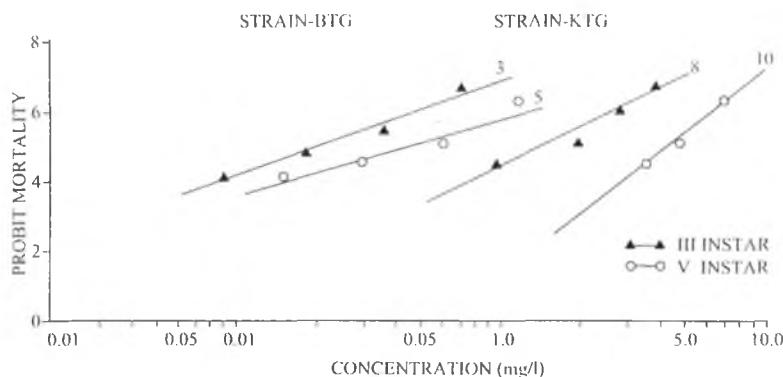


Fig. 2: Showing log concentration-probit regression lines of phosphine for different instars of larvae of susceptible and resistant strains of *T. granarium*

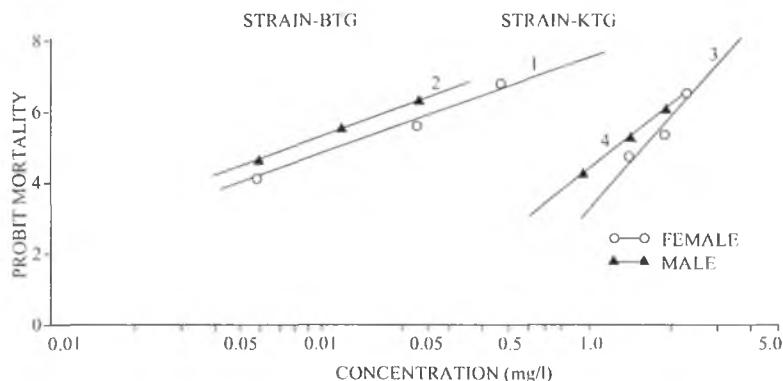


Fig. 3: Showing log concentration-probit regression lines of phosphine for adults of susceptible and resistant strains of *T. granarium*

gression line of phosphine for third-instar larvae of *T. granarium* increased from 2.09 to 3.21 (Fig. 2) as a result of selection, however, the slope for fifth-instar larvae was decreased from 7.44 to 6.22. The flattening of slope of log-concentration probit regression line for adults (males and females) was also observed (Fig. 3). In general, the flattening of slopes of log-concentration probit regression lines through selections for insecticides resistance has been observed (Brown and Pal, 1971). The increase in slope may, therefore, be either merely a reduction in variability of the strain with respect to its response to phosphine or the polygenic nature of phosphine resistance. However, no information on the inheritance of phosphine resistance in insects is available.

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# Ultrastructure of the Female Accessory Reproductive Gland of *Gesonula punctifrons* (Orthoptera: Acrididae) in relation to its Secretion

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**Abstract:** The female accessory gland of late vitellogenic grasshopper (*Gesonula punctifrons*) has been analyzed at the ultrastructural level. It liberates both lipoidal and proteinaceous secretion within the lumen of the accessory glands which ultimately are discharged along with the eggs. The accessory gland cells are columnar in nature and contains rich supply of Golgi vesicles and mitochondria. The cells also contain profuse supply of free ribosomes and rough endoplasmic reticulum. The secretion may play vital role in protection of the egg as well as fertilization.

**Keywords:** *Gesonula punctifrons*, female accessory reproductive gland, ultrastructure, secretion.

## INTRODUCTION

The accessory reproductive glands of the insects vary greatly in structure, location and secretory products (Rockstein, 1964). The ultrastructural study of the maturation of female accessory reproductive glands complex of *Schistocerca gregaria* has revealed the presence of the secretion in the cytoplasm of the cell having free ribosomes, smooth surfaced ER elements; few small and inactive Golgi complexes and abundant mitochondria (Odhiambo 1969, 1971). It has also been reported by same authors that the chromatin of the accessory gland cells is in more dispersed state indicating some organizational changes at the level of the nucleus. In Acrididae these gland cells have been shown to be mesodermal in origin and open into the vestibulum. But the proper understanding of the process of secretion by the female accessory gland is far from clear. The secretions have also not been characterized. It has also been noted that studies on structural and physiological nature of the female accessory gland are comparatively few and secretory dynamics of the female accessory gland on a wide scale need careful analysis while compared to that of male accessory reproductive gland. In view to

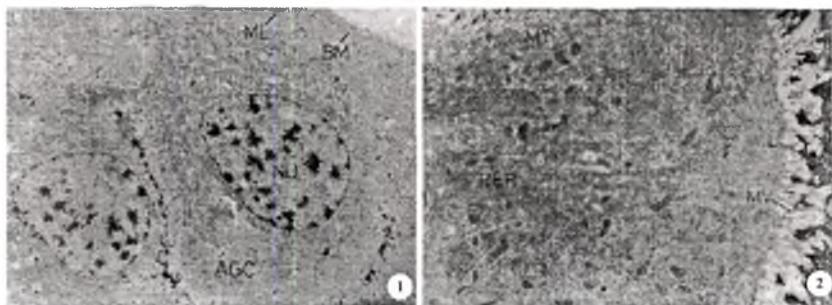


Fig. 1: Electron micrograph of accessory reproductive gland showing the accessory gland cell (ACG) resting on a basement membrane (BM). The outer thin connective tissue layer (ML) is also seen.  $\times 1500$

Fig. 2: EM view of cytoplasm of the accessory reproductive gland cell opening in the lumen (L) of the gland as microvilli (MV). Adi-electronic droplets are present in the lumen and the intercellular space. Electron dense secretion present in mitochondria (arrow) and RER are shown.  $\times 4500$

above, the present work reports the fine structural details of the process of secretion by the female accessory reproductive gland of *Gesonula punctifrons*, a common rice field grasshopper in Tripura and West Bengal.

#### MATERIAL AND METHODS

Accessory reproductive glands from the adult female *G. punctifrons* were dissected out in Insect Ringer solution. The tubular structure was then washed in Millong's buffer (pH 7.2) containing 0.1 M  $\text{CaCl}_2$ . Washed parts were fixed in 5% glutaraldehyde for 2–3 hours at room temperature. The fixed tissues were again washed with a number of changes of buffer for 2–3 hours at room temperature. Then the tissues were osmicated for 1 hour and subsequently washed in distilled water. Embedding of the tissues were done using epoxy resin CY212, hardener HY964 and accelerator DY064 with different gradation. Finally the tissues were embedded in a template at 60°C for 48 hours.

Ultrathin sections were cut on a LKB ultratome with glass knives and stained with uranyl acetate and lead citrate (Reynolds, 1963) and viewed under a transmission electron microscope on naked copper grids. Photographic enlargements were made and printed on glossy papers for analysis.

#### RESULTS

The cellular body of the accessory reproductive gland is formed by a single-layer of columnar epithelial cells juxtaposed with one another which rest on a basement membrane that is about 300 nm in thickness and is covered by a thin layer (4  $\mu\text{m}$ ) of connective tissue layer (Fig. 1). The connective tissue layer is reinforced by muscle cells having myofibrils and the plasma membrane of these cells vary from region to region. Septate desmosomes, about 40 nm thick are observed near the apical lateral region

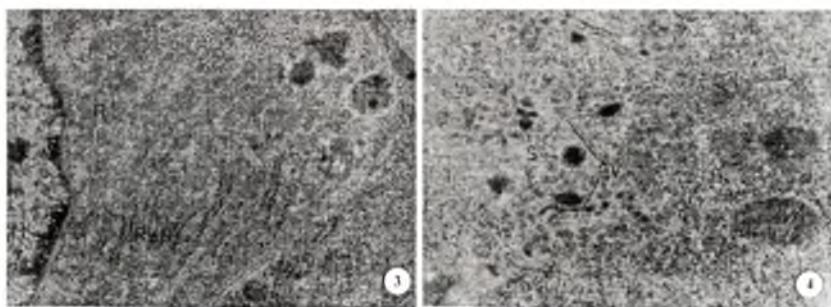


Fig. 3: Highly magnified electron micrograph of the accessory reproductive gland cell showing RER, free ribosome (R), microtubules (M) and nuclear heterochromatin (H).  $\times 18000$

Fig. 4: Magnified view of the Golgi complex (GV) in the accessory reproductive gland cell. Golgi bodies giving rise to secretory droplets (S) are apparent. Mitochondria (MT) with cristae are also visible.  $\times 24000$

and the luminal surface consists of microvilli (length about  $1\ \mu\text{m}$ , diameter 200 nm). The microvilli of neighbouring cell border forms a minute canalliculi or crypt in the wall of the accessory gland (Figs. 2 and 4). These crypt contains combination of electron dense and electron lucent granules of ovoid shape and their opacity indicates their combined nature of lipids and proteins.

The nuclei of the epithelial cells are seen with some pores containing RNP particles in their vicinity (Fig. 3). Heterochromatic portion is attached to the inner surface of nuclear envelope as dense masses and euchromatic portion remains as finely granular mass dispersed in the nucleus. The cytoplasm of these cells is heterogenous in organization and could be distinguished into basal and apical parts (Figs. 2, 3 and 4). The basal part possesses many RER, free ribosomes, secretory droplets and generally elongated mitochondria (diameter about  $2.5\ \mu\text{m}$ ) with distinct cristae and other membrane bound vesicles with or without any content (Figs. 2, 4 and 5). The RER cavities contain moderately electron dense granular materials (Fig. 3) and ribosomes (diameter about 25 nm) are attached to the membrane. Some mitochondria are observed to contain electron dense secretory material. Golgi apparatus with highly developed cisternae are observed in this region to give rise to the secretory materials (Fig. 2). These secretory bodies/droplets have no membrane. Further, microtubules (diameter 25 nm) are also observed in this region (Fig. 3).

In the apical and subapical regions, abundant RER lie parallel to the lateral plasma membrane and polyribosomes are evenly distributed (Figs. 2 and 5). Electron dense secretory materials and several profiles of microtubules (width about 25 nm) are also present in this region (Figs. 1 and 5). Tip of this region appears with microvilli having less dense cytoplasm and no cell organelle (Fig. 5). Well-defined highly electron lucent granules (300–400) nm are present in this region along with mitochondria and ribosomes which appear even upto the base of the microvilli (Figs. 2 and 5). The most important feature of these cells are presence of huge net work of Dictyosomes or Golgi vesicles, (Fig. 4). The Golgi complexes are actively engaged in secretion as could be

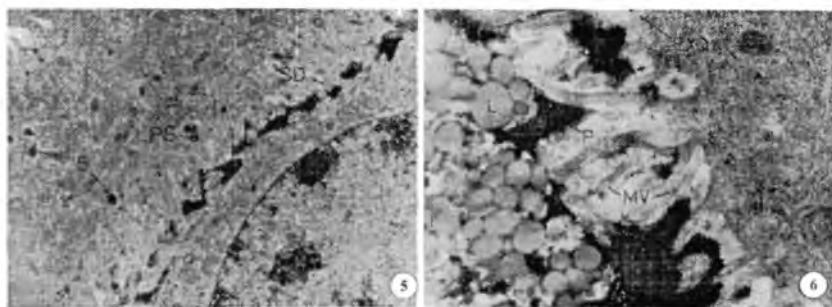


Fig. 5: Electron micrograph showing the intercellular crypt in between two accessory gland cell showing electron dense and electron opaque droplets (SD). Intercellular secretory products (S) and lipoprotein secretions (LPS) are also shown.  $\times 6000$

Fig. 6: Magnified view of the apical cytoplasm of the accessory reproductive gland cell showing ramified microvilli and luminal lipoid (L) and protein (P) secretions. The exocytotic pockets (EXO) are also shown.  $\times 18000$

evidenced by the presence of secretory vesicles (Fig. 4). The outer lining of plasma membrane is not very clear. Two types of secretory materials, electron opaque droplet (diameter 200–500) nm and cluster of electron dense matrix are observed within the lumen of the gland even at the spaces between the microvilli. These indicate that the secretion consists both of proteins and lipids. In the vicinity of the outer margin of the epithelium signs of exocytosis are very clear (Fig. 6). Proteinaceous secretions are seen near these spots of exocytosis. Both within the cellular crypt and the lumen of the accessory gland both the secretions (lipid and protein) are discharged. Some lipoprotein granules are seen to be liberated within the crypts (Fig. 5). In the tubular part of the gland the secretory products are seen to be stored. These secretions retain their identity. From these EM pictures it is abundantly clear that this total secretion is discharged alongwith eggs during the time of parturition.

## DISCUSSION

The results indicate that the female accessory glands of the grasshopper *Gesonula punctifrons* are active in synthesis and liberation of lipoprotein secretion. These secretions are elaborated in the lumen of the accessory glands by the process of exocytosis. The scheme of elaboration and discharge of secretory products is probably very close to that outlined in mammalian secretory cells (Palade and Siekevitz, 1956; Caro and Palade, 1964). Microtubules are possibly associated with the movement of secretory product (Fawcett, 1979). Similar to the findings of Lanzavecchia (1964), some well-formed mitochondria are observed to accumulate the products during the process of synthesis of the secretory droplets. The analysis of the secretory droplets is suggested to be lipoprotein in nature. Fausto *et al.* (1997) have undertaken similar study on *Phlebotomus perniciosus* and they have highlighted the similar process of exocytosis. They have identified the secretion to be proteinaceous. But in the present study it has been

revealed that the secretion is lipoprotein in nature.

Szopa (1981a) has discussed the role of accessory reproductive glands and genital ducts in egg pod formation in female *S. gregaria*. She has shown that the removal of accessory gland does not prevent the formation of egg plugs and hence in an extension of oviduct. Further, Szopa (1981b) has established that the maturation and secretion of the accessory glands is dependent on the activity of the corpus allatum and is controlled by the juvenile hormone. From morphological studies of the accessory gland it may be stated that in *Gesonula punctifrons*, the same type of mechanism operates. However, extensive biochemical studies are very much required to further understand the nature of secretion as well as the process of its elaboration.

#### ACKNOWLEDGEMENTS

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## Chemical Analysis of Secretion from Abdominal Scent Glands of Nymphal *Cyclopelta Siccifolia* Westwood (Hemiptera: Pentatomidae)

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**Abstract:** Chemical analysis of the nymphal exocrine secretion of *Cyclopelta siccifolia* Westwood by gas chromatography and mass spectra showed a blend of seven compounds, Diisopropanol amine, n-butyl butyrate, o-isopropenyl aniline, trans-dec-2-enal, Ethyl myristate, octadecyl amine and  $\alpha$ -(trichloro methyl) benzyl acetate. These volatile compounds are irritative, corrosive and are likely to provide defence against predators.

**Keywords:** *C. siccifolia* nymph, abdominal scent glands, defence secretion.

### INTRODUCTION

Adults and nymphs of pentatomid bugs are notorious for the production of noxious exocrine secretions from the scent glands whenever disturbed or irritated. These secretions are highly volatile and are usually emitted from the scent glands as droplets or are sprayed at or injected into potential predators (Valocurone Dazzine and Vita Finzi, 1974). Pentatomid defence secretions are composed of a dazzling array of complicated chemical compounds (Aldrich *et al.*, 1984; Staddon and Olagbemiro, 1984; Blum, 1985; Gough *et al.*, 1985; Aldrich, 1988; Surender and Janaiah, 1990; Janaiah, 1993; Vidyasagar, 1995; Srinivasulu *et al.*, 1996).

The present investigation deals with the structure and chemical analysis of the abdominal scent glands of nymphs of the pentatomid phytophagous bug, *Cyclopelta siccifolia*.

### MATERIAL AND METHODS

Nymphs of *Cyclopelta siccifolia* (Westwood) were collected from the host plant, *Sesbania speciosa* (Fabaceae) in the betel gardens, at Bhuvanagiri (A.P) and from the intermediate host plant, *Pongamia pinnata* (Fabaceae). They were maintained in the

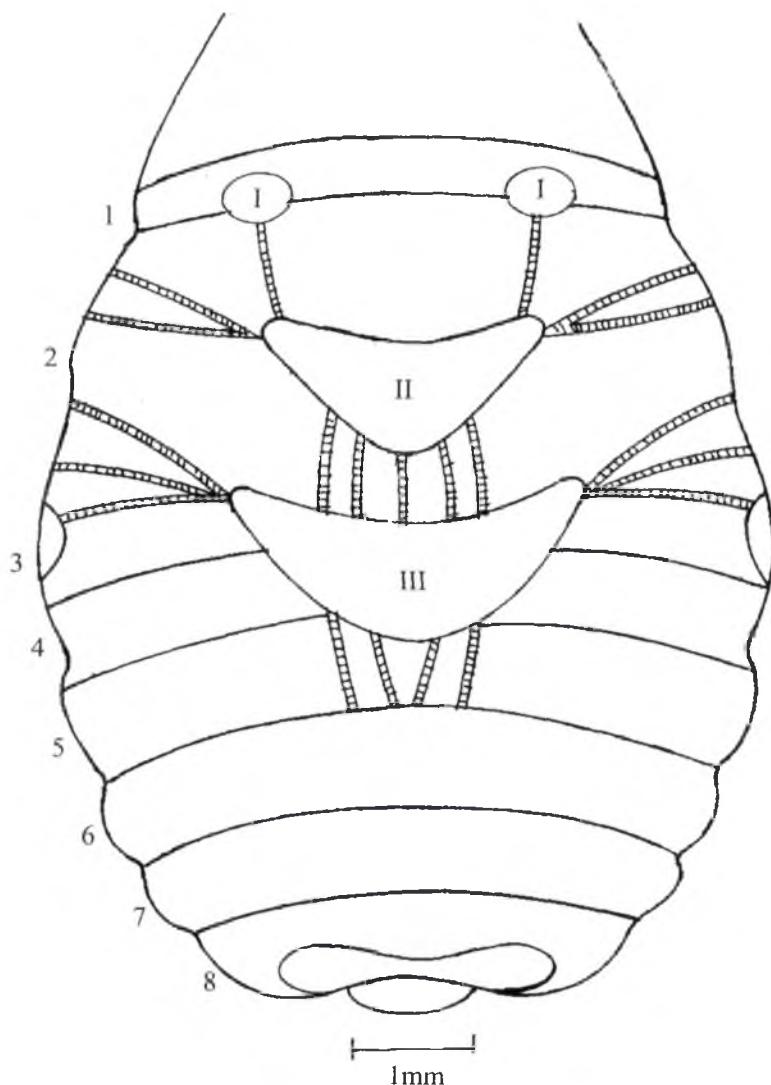


Fig. 1: Nymphal abdominal scent glands of *C. siccifolia* I, II and III are abdominal secent glands with associated regulatory muscles.

laboratory on the cut leaves of host plant at room temperature for a week. The scent secretion was collected from the second and third pair of abdominal scent glands of 50 nymphs by placing microcapillaries against the gland openings and pooled.

The pooled secretions were analysed by gas chromatography and mass spectrometry (GC-MS) and the components were identified with the help of authentic samples obtained from Aldrich Chemical Company, Milwaukie, U.S.A.

GC-MS analysis was conducted using a Finnigan MAT quadrupole 1020B Mass

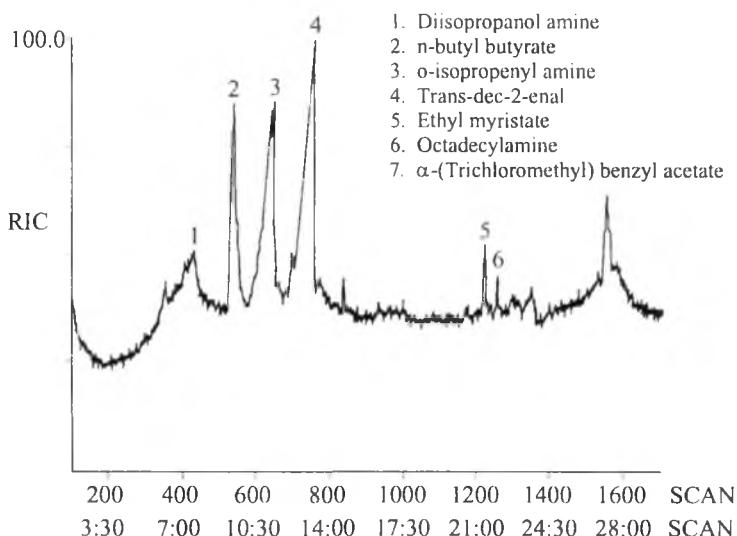


Fig. 2: Gas-Chromatographic separation of abdominal secent gland secretions of *C. siccifolia* nymphs.

Spectrometer with Hewlett Packard high performance capillary column crosslinked methyl silicane film of thickness 0.52 microns, and internal diameter of 0.32 mm and the flow of helium was 2 ml/minute.

The samples (unknown and authentic) of 0.3  $\mu$ l to 0.5  $\mu$ l were injected through sample boat separately and the column was held at 60°C for one minute. The samples were programmed for 50–200°C for 30 minutes. All the spectra were run at 70 ev at manifold temperature of 70°C.

## RESULTS

Three pairs of abdominal scent glands are present in all the five nymphoid instars of *C. siccifolia*. These glands each with a pair of ostioles are situated dorsally in a row on the abdominal segments 1 to 5 (Fig. 1). The first pair of glands are small, brick red, and oval, while the second and third pairs are large and crescent shaped. These glands are provided with regulatory muscles.

The GC-MS analysis of the scent secretion of II and III pair of scent glands showed a chromatogram consisting of mainly seven peaks (Fig. 2) each represented a chemical compound (Table 1). Peak-1 showed a mass spectrum with molecular ion( $M^+$ ) of low intensity at m/z 133 and base peak appeared at 43. The compound was identified as Diisopropanol amine. Peak-2 showed the mass spectrum with low intensity of  $M^+$  at m/z 144 and base peak appeared at m/z 43. This gave evidence that the compound was n-butyl butyrate. The mass spectrum of peak 3 showed  $M^+$  at m/z 133 with the base peak at m/z 57. The compound was confirmed as o-isopropenyl aniline. Mass spectrum of peak-4 with molecular ion at m/z 154 and base peak at m/z 41 identified

Table 1: Chemical components of the abdominal scent gland secretions of *Cyclopelta siccifolia* nymphs with GC-MS evidence.

Peak No.	Component	Molecular Weight	Masses of ions in order of abundance
1.	Diisopropanolamine	133	43 (Base peak), 44, 61, 71, 75, 89, 116, 133 (Molecular ion).
2.*	n-butyl butyrate	144	43 (Base peak), 44, 55, 72, 84, 101, 144 (Molecular ion).
3.	o-isopropenyl aniline	133	45, 57 (Base peak), 73, 84, 101, 133 (Molecular ion).
4.*	<i>trans</i> -dec-2-enal	154	41 (Base peak), 55, 73, 82, 99, 110, 154 (Molecular ion).
5.*	Ethyl myristate	256	44, 55 (Base peak), 59, 67, 77, 89, 106, 120, 133, 148, 163, 198, 256 (Molecular ion).
6.	Octa decylamine	269	43 (Base peak), 47, 73, 89, 95, 133, 155, 180, 243, 361 and 269 (Molecular ion).
7.	$\alpha$ -(trichloromethyl) Benzylacetate	267	41, 43 (Base peak) 115, 129, 185, 196, 267 (Molecular ion).

\*Compared with authentic samples

as *trans*-dec-2-enal. The other peaks 5,6 and 7 were identified as Ethyl myristate, Octadecyl amine, and  $\alpha$ -(Trichloromethyl) benzyl acetate.

## DISCUSSION

Whenever nymphs of *C. siccifolia* are disturbed or irritated a characteristic odour is usually produced. The scent secretions is highly pungent and suffocative, causing irritation of the mucous membrane of nostrils. Birds can be repelled by volatile irritants from the pentatomids (Pasteels *et al.*, 1983). In our observation, the house sparrow, *Passer domesticus* on seeing the nymphs of *C. siccifolia*, suddenly flew down and stretched it's neck, perhaps to pick the nymphs but it got frightened and immediately retracted it's neck and flew swiftly into the sky. Perhaps, this is due to the release of pungent scent by the nymphs appeared to have hit the eyes, to encounter the danger. The pungency is mostly due to the presence of aldehyde, *trans*-dec-2-enal, and probably the major contributor to the odour of the nymph of *C. siccifolia*. When the nymphs of *Halys dentatus* were offered to garden lizards and wood-peckers they first swallowed one or two but regurgitated later (Srinivasulu *et al.*, 1996).

When the secretions from the nymphs of *C. siccifolia* is spilled on the human skin, they caused irritation and burning sensation followed by formation of blisters which remained for few hours and left a yellow stain on the skin. The scent component responsible for corrosiveness is predominantly *trans*-dec-2-enal. Similar observations were noticed with the scent secretions from the metathoracic scent glands of *Tessaratoma javanica* (Janaiah, 1993), abdominal scent secretions of *Libyaspis angolensis* (Cmelik, 1969), metathoracic scent glands of *Biprorulus bibax* (Macleod *et al.*, 1975) and *Nezara viridula* (Gilby and Waterhouse, 1965).

In *Dysdercus intermedius* (Youdeowei and Calam, 1969), it was observed that first and second abdominal scent gland secretions were responsible for aggregation of nymphs while the secretions from the third abdominal scent glands appeared to have a defence role. The secretions from the second and third pair of abdominal glands of nymphs of *C. siccifolia* indicated mainly defensive role protecting the larvae from predators (Vidyasagar, 1995). Ethyl myristate, likely to be important ester in nymphs of *C. siccifolia* was reported earlier from the tarnished plant bug, *Lygus lineolaris* (Gueldner and Parrott, 1978).

Diisopropanol amine and o-isopropenyl aniline have not, as far we know, been previously reported in any pentatomid bugs. Diisopropanol amine and octadecyl amine were responsible for corrosiveness and o-isopropenyl aniline caused irritation on the lips and skin of humans (Vidyasagar, 1995). n-butyl butyrate (BB) was also earlier reported from the scent secretion of adult coreoid bugs *Amorbus rhombifer* (Waterhouse and Gilby, 1964) and from the nymphal glands of *Cyrysocoris purpureus* (Janaiah *et al.*, 1988). BB acts as wetting agent to facilitate the penetration of main toxicant into the host body (Blum, 1978). In addition, BB is a potent fungicide at higher concentrations (3.30%) causing total spore germination inhibition of *Dreschlera specifera* and *Fusarium oxysporum* (Surender *et al.*, 1987).

The nymphal abdominal scent secretions of *C. siccifolia* are enriched with high molecular weight constituents, presumably because the wingless nymphs require longer lasting protection (Srinivasulu *et al.*, 1996). We conclude in the nymphs of *C. siccifolia*, all the components of scent secretions are primarily defensive in nature.

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## Immature Stages of Two Orthoclads (Diptera: Chironomidae) of Darjeeling Himalayas

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**Abstract:** Larvae and pupae of two orthoclads, *Cricotopus* (*Cricotopus*) *pyrus* Chaudhuri and Ghosh and *Orthocladius* (*Eudactylocladius*) *androgynus* Bhattacharya *et al.* are described from Darjeeling Himalayas of West Bengal. The species were previously known from Bhutan and India respectively.

**Keywords:** Immatures, orthoclads, Darjeeling–Himalayas

The orthoclads are the largest group of chironomid midges occupying the widest range of habitats. The larvae are primarily cold adapted living in all types of lentic, lotic, semiterrestrial to terrestrial habitats. They decrease in number in increasingly warmer regions, though they are not uncommon in many warmer regions (Fittkau, 1964; Chattapadhyay and Chaudhuri, 1991). In general, they become more common towards higher altitude or with increasing altitude and are poorly represented in tropical water (Ashe *et al.*, 1987). Orthocladid midges in adults of the genera *Cricotopus* v.d. Wulp and *Orthocladius* v.d. Wulp had been described in 8 and 9 species respectively but the immatures of any of the two genera remained unknown in India before this investigation (Sublette and Sublette, 1973; Chaudhuri and Ghosh, 1982; Chaudhuri and Guha, 1987; Bhattacharya *et al.*, 1991).

Larvae were obtained from lotic situations with the aid of nylobolt plankton net of 30 meshes/cm<sup>2</sup> and those inhabiting in larval cases were taken out of the surface with the help of scalpel and small forceps. The material were then carried to the laboratory in vacuum containers filled with water from their environment following Pinder (1983). The larvae were reared after Epler (1992) by putting them in shallow petridishes of 5 cc and or 12 cm in diameter with little amount substrates like macrophytes plus a moderate amount of water. The experimental dishes were then placed in individual small cages, aerated by electric air pumps and kept at a temperature somewhat above that of the habitats. A small amount of food in the form of algal filament or mosses or leaves of aquatic macrophytes were added twice a week. Development of the worms were examined periodically in order to recognise prepupal larvae. The prepupae were transferred to individual rearing vials of different sizes containing little water plugged with nonabsorbent cotton. Newly emerged imagines in separate vials were kept in

cool dark place for 1 or 2 days allowing sclerotization. Specimens were suitably slide-mounted following Pinder (1989) for microscopical study.

Terminologies and usages as recommended by Saether (1980, 1989) have been followed here.

Measurements are in millimeter (mm) with the number before parantheses denoting average value while those within the brackets indicate minimum-maximum suffixed by "n" being the number of specimen encountered.

Specimens of the present study are with the collections of insects of the Entomology Laboratory, Department of Zoology, University of Burdwan and will be deposited to the National Zoological collections (NZC), Calcutta in time.

#### ***Cricotopus pyrus Chaudhuri and Ghosh***

*Cricotopus (Cricotopus) pyrus Chaudhuri and Ghosh, 1985: 45; Chaudhuri and Guha, 1987: 26.*

##### *Fourth instar larva:*

Body yellowish brown; head capsule yellowish; claws of anterior and posterior parapods yellow and yellowish brown respectively. Exuviae transparent. Total body length 4.94 (4.53–5.34, n = 3).

**Head:** Mandible dark brown, premandible yellowish brown, mentum dark brown, occipital margin light brown. Ventral head capsule (Fig. 1J) 0.450 (0.420–0.480, n = 3) long and 0.310 (0.300–0.330, n = 4) wide. Eye spot paired, black and dorsal one shorter than the ventral one.

**Antenna** (Fig. 1A): 5 segmented, length ratio of antennal segments (I–V): 12 : 4 : 1.25 : 1.25 : 1. AR 1.6. Basal antennal segment 0.013 wide with a ring organ 0.003 in diameter, distance from base to ring organ 0.007. Blade 0.027 long. Lauterborn organs 0.005 long; style 0.009 long, longer than the length of 3rd antennal segment. **Labrum** S I (Fig. 1B): bifid, S II (Fig. 1B) long, slender and simple, other S setae simple. Chaetae serrate. Pectin epipharyngis consisting of 3 scales, median scale subequal to lateral scales. Premandible (Fig. 1C) 0.068 long with single tooth, brush very weakly present. **Mandible** (Fig. 1D): 0.135 (0.120–0.160, n = 3) long with one apical, 3 inner and one false tooth, mandible apically dark brown and basally yellow. Seta subdentalis 0.018 long, apically bluntly pointed. Seta interna with 5–6 simple branches. Outer margin of mandible very little crenulate. **Mentum** (Fig. 1E): With flattened width 0.102 (0.099–0.110, n = 3), median tooth 0.021 wide. Ventromental plate (Fig. 1E) weak, 0.068 wide. **Maxilla** (Fig. 1F): Palpiger with minute, triangular chaetulae. Setae maxillaris present, simple.

**Body** (Fig. 1G): Yellow. Procercus 0.016 high and 0.011 wide bearing 4–6 setae and with longest seta being 0.416 long. Supra anal seta 0.060 long. Sa/An 0.142. Posterior parapods 0.24 with 14–16 yellow coloured variable claws (Fig. 1H). Anal tubules 4, finger-like, 0.204 long, slightly shorter than length of posterior parapods. Abdominal segments V–IX each with one pair of setal tufts (Fig. 1I).

**Pupa:** Exuviae brown. Total body length 3.12 in male. Th/AM 1.22.

**Cephalothorax:** Frontal seta absent; frontal warts absent; frontal apotome wrinkled (Fig. 2A). Antennal sheath (Fig. 2C) 0.610 long. Median antepronotals 0.096 and 0.011 long; lateral ante-pronotal 0.080 long. Thoracic horn elongate, with spines

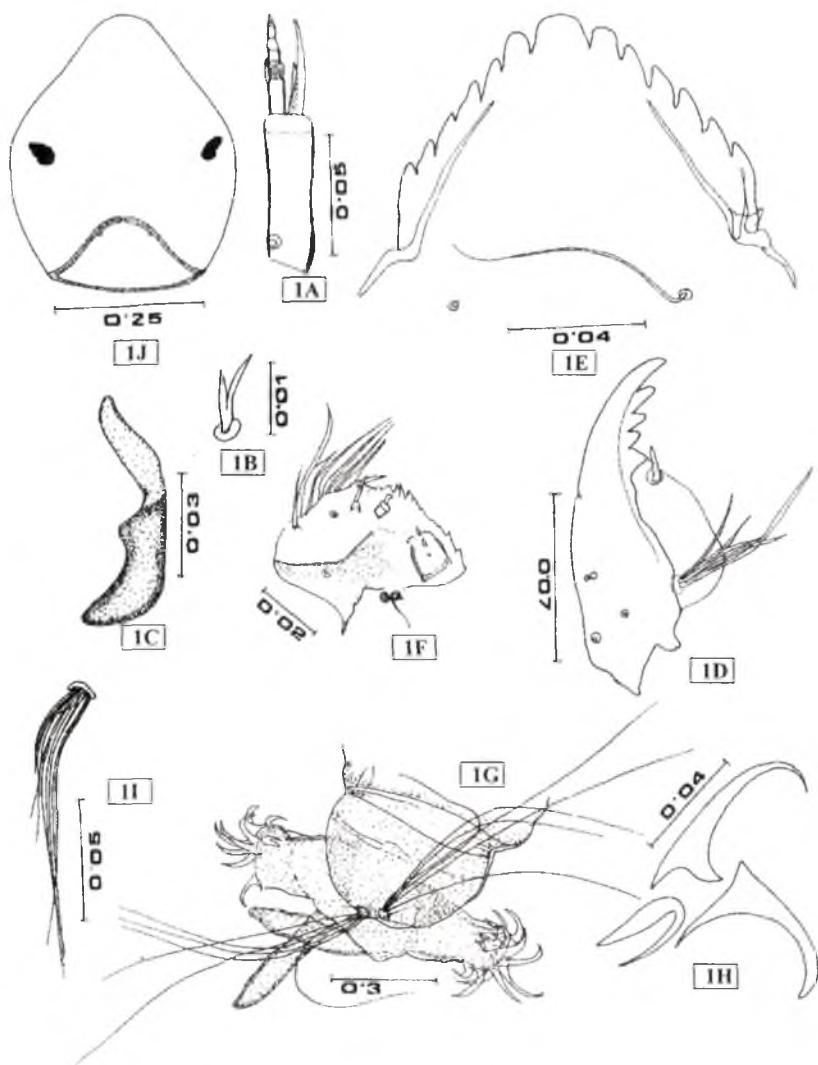


Fig. 1: A–J: Larva of *Cricotopus pyrus* Chaudhuri and Ghosh. A. Antenna; B. S I; C. Pre-mandible; D. Mandible; E. Mentum and ventromental plate; F. Maxilla; G. Posterior abdominal segments; H. Claws of posterior parapods; I. Abdominal setal tuft and J. Head capsule (ventral view).

(Fig. 2B). ThR 3.50. Anterior precorneal seta 0.120, median seta 0.112 and posterior seta 0.128 long (Fig. 2B); dorsocentrals arranged in 2 pairs; Dc<sub>1</sub> 0.028, Dc<sub>2</sub> broken, Dc<sub>3</sub> 0.052 and Dc<sub>4</sub> 0.020 long; distance between Dc<sub>1</sub> and Dc<sub>2</sub> 0.032, between Dc<sub>2</sub> and Dc<sub>3</sub> 0.116 and between Dc<sub>3</sub> and Dc<sub>4</sub> 0.020. Wing sheath 0.720 long.

**Abdomen** (Fig. 2D): Tergites I–III without shagreen; IV and V with shagreen restricted on anterolateral region; VI with shagreen on anterior half; VII with shagreen

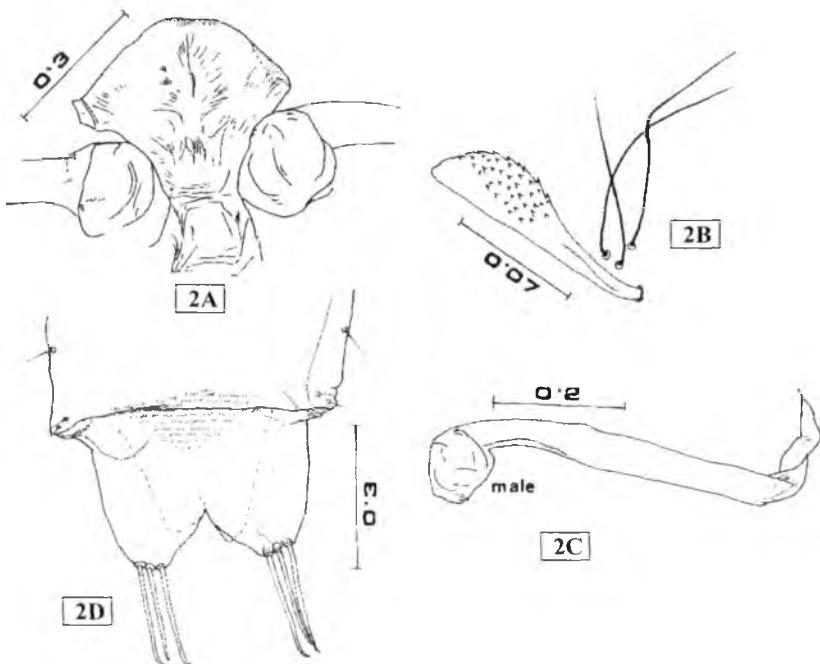


Fig. 2: A-D: Pupa of *Cricotopus pyrus* Chaudhuri and Ghosh. A. Frontal apotome with prefrons; B. Thoracic horn and precorneal setae; C. Antennal sheath (male) and D. Tergite IX with a portion of T VIII.

divided into anterior end posterior fields and covering nearly entirely on VIII; IX with anterior band of shagreen. T III to VI with anteriorly directed two median, oval, spine patches in close approximation and 2-3 rows of anteriorly directed spines at posterior end, fuses partially at lateral extremity leaving a narrow bare gap of with the median patches. T II with 2 rows of dark brown coloured hooklets. Conjunctives III-V with anteriorly directed spinules. Sternites I-II without shagreen, III-VIII with shagreen. Pedes spurii B large, bulbous, present on segment II. Pedes spurii A present on segments IV-VII, consisting of broad patch of anteriorly directed spinules. Apophyses absent.

Anal lobe (Fig. 2D) semicircular, 0.184 long, 0.208 wide, bearing three apically hooked macrosetae of which middle one is shorter than others, of 0.116, 0.104 and 0.116 long. Genital sac in male slightly overreaching anal lobe; G/F 1.086. ALR 1.76.

#### *Material examined*

2 larvae, West Bengal, Ghum ( $27^{\circ} 02'N$  Lat.,  $88^{\circ} 16'E$  Long., Alt. 2256 m msl), 14.VII. 91, Coll. D. K. Som; 1 male with pupal and larval exuviae (reared), data same as larvae; *Holotype* male, (Type No. B.U. Ent. 97), Bhutan, Thimpu ( $27^{\circ} 32'N$  Lat.,  $89^{\circ} 53'E$  Long., Alt. 2012 m msl), 10.X.78, Coll. Dr. B. C. Nandi.

Table 1: Setae present on segment I–VIII

	I	II	III	IV	V	VI	VII	VIII
Dorsal (D)	2	4	4	4	4	4	4	2
Lateral (L)	2	3	3	3	3	3	3	4

### Remarks

Chaudhuri and Ghosh (1985) described the adult male of *Cricotopus (Cricotopus) pyrus* from Bhutan, adjacent to the eastern border of Darjeeling montane area. Most of the morphological and morphometric features viz., arrangement of setae on abdominal tergites, AR value of less than 1.00, shape of inferior volsella and higher BV of mid and hind legs of adult male, absence of frontal setae, subequal anal macrosetae, absence of strong shagreen on tergites VII and VIII, widely separated anterior and posterior tergal shagreen, short L<sub>4</sub> of segment VIII of pupa, simple premandible, first and second lateral teeth of mentum not partially fused with median tooth and with a only few distally pinnate lamellae of galea of larva speak in favour of placing this species in the *bicinctus*-species group of Hirvenoja under subgenus *Cricotopus*. However, morphometric values of larval mentum of the present species differ a little from those of other members of the group. Similarly, in case of pupa, pedes spurii B is found to be visible only on segment II in the present species instead of segments II and III as noted in others of the same group. The possession of the following characters of larva, pupa and adult male place it as a member of the *bicinctus*-group of the subgenus *Cricotopus*:

*Adult:* i) Very low AR value of 0.37, ii) mesonotum brown with three darker vittae, iii) colour pattern of abdominal tergites, iv) pyriform inferior volsella and v) sharply elevated crista dorsalis.

*Pupa:* i) Characteristic shagreen pattern of tergites, ii) pedes spurii B large, bulbous, present only on segment II and iii) pedes spurii A present on segment IV–VII.

*Larva:* i) AR 1.6, ii) premandible with very weak brush and iii) abdominal segments V–IX each with one pair of long (0.160–0.170) setal tufts.

### *Orthocladius androgynus* Bhattacharya *et al.*

*Orthocladius (Eudactylocladius) androgynus* Bhattacharya *et al.* (1991, 338).

#### *Fourth instar larva:*

Body colour yellow; exuviae transparant; head capsule brown; claws of anterior and posterior parapods yellow and brown respectively. Total body length 6.11 (5.82–6.66,  $n = 3$ ).

*Head:* Mandible, premandible, mentum and occipital margin dark brown. Ventral head capsule (Fig. 3A) 0.440 (0.420–0.460,  $n = 3$ ) long and 0.330 (0.328–0.332,  $n = 3$ ) wide. *Antenna* (Fig. 3B): 5 segmented, length ratio of antennal segments (I–V): 12 : 3 : 12 : 1.5 : 1. AR 1.79. Basal antennal segment 0.014 wide with a ring organ of 0.005 in diameter, distance from base to ring organ 0.013. Blade 0.017 long. Lauterborn organs 0.003 long. *Labrum:* S I (Fig. 3C) bifid, other S setae simple. Chaetae with simple long branches. Spinulae simple. Pecten epipharyngis with 3 subequal scales,

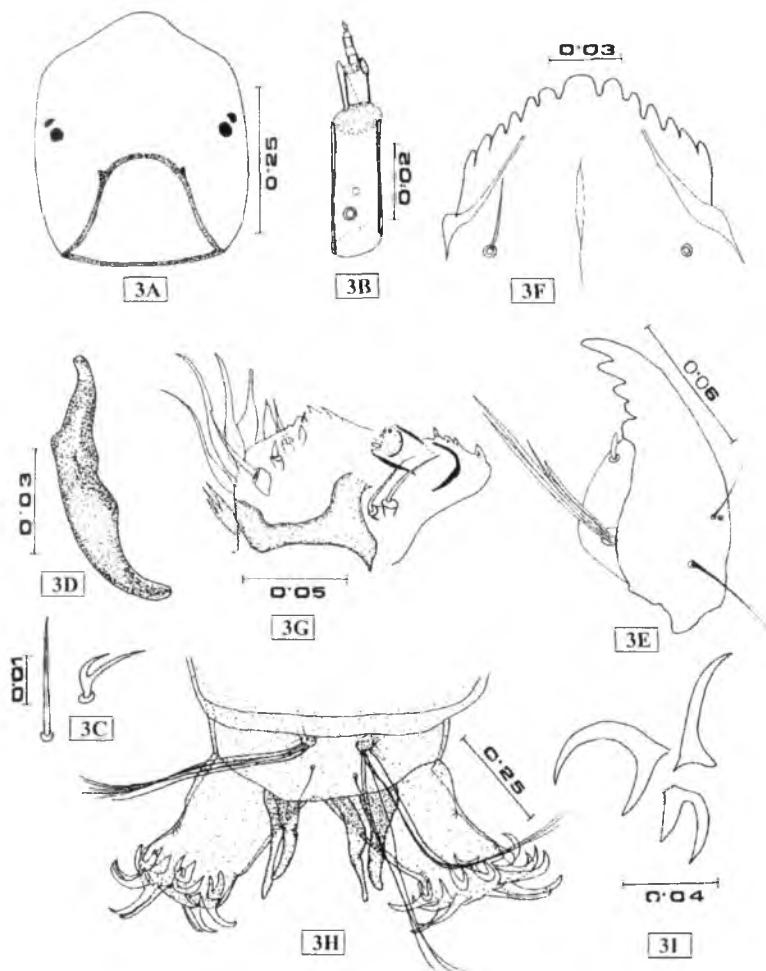


Fig. 3: A-I: Larva of *Orthocladius androgynus* Bhattacharyay, Ali and Chaudhuri. A. Head capsule (ventral view); B. Antenna; C. S I and S II; D. Premandible; E. Mandible; F. Mentum and ventromental plate; G. Maxilla; H. Posterior abdominal segments and I. Claws of posterior parapods

middle one broader than lateral scales. Premandible (Fig. 3D) 0.076 long, with single tooth and with or without brush. *Mandible* (Fig. 3E): 0.137 long with 1 apical and 3 inner teeth and 1 false tooth; apical tooth not longer than combined width of 3 inner teeth; mandible dark brown apically and pale brown basally. Seta subdental is 0.014 long, pointed. Seta interna with 4 simple branches. *Mentum* (Fig. 3F): With flattened width 0.094, median tooth 0.016 wide. MR 1.88 (1.60–2.00,  $n = 4$ ). Ventro-mental plate (Fig. 3F) 0.061 wide, moderately developed and extended beyond outermost lateral tooth. *Maxilla* (Fig. 3G): With

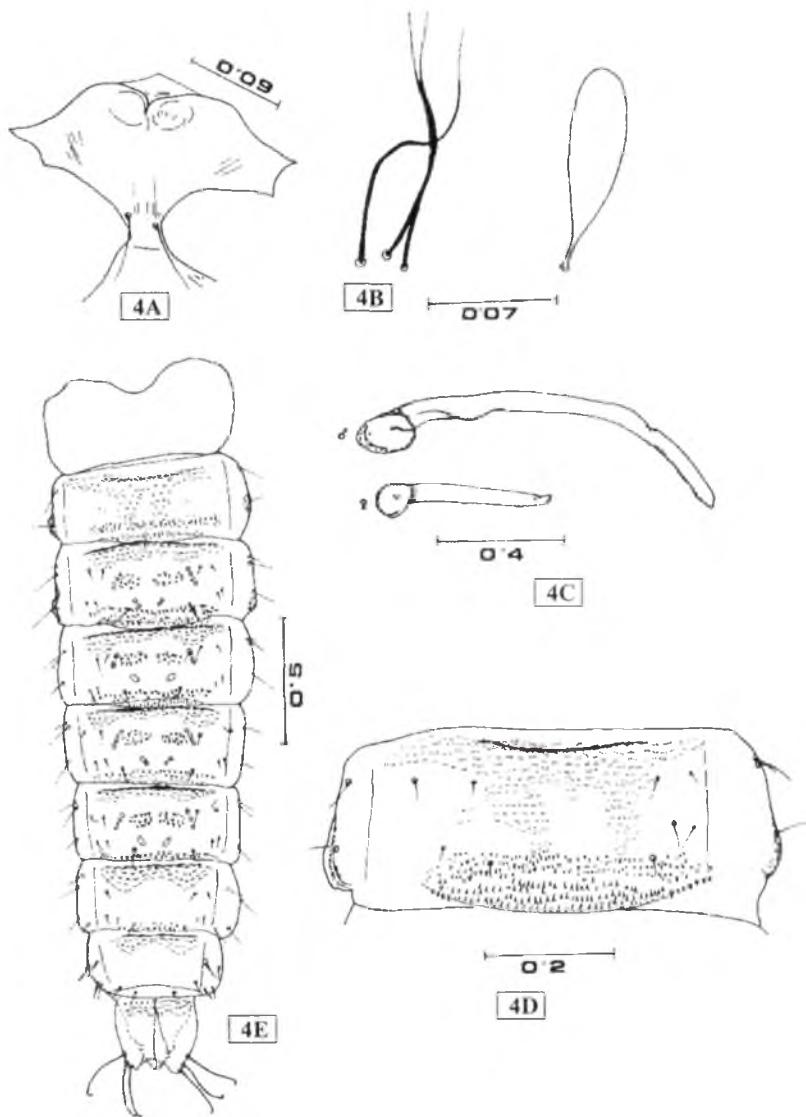


Fig. 4: A–E: Pupa of *Orthocladius androgynus* Bhattacharyay, Ali and Chaudhuri. A. Frontal apotome with prefrons; B. Thoracic horn and precorneal setae; C. Antennal sheath (male and female); D. Tergite II and E. Abdomen (tergites)

simple, triangular chaetulae of palpiger. Galea with 4–5 simple lamellae. Anterior lacinial chaeta leaf like. Pecten galearis absent.

*Body* (Fig. 3H): Light yellow. Procerus 0.021 high and 0.025 wide, bearing 6 anal setae and with longest anal seta 0.320–0.340 long. Supraanal seta 0.057 long. Sa/An 0.17. Posterior parapods 0.221–0.238 long with 16–17 sclerotised variable

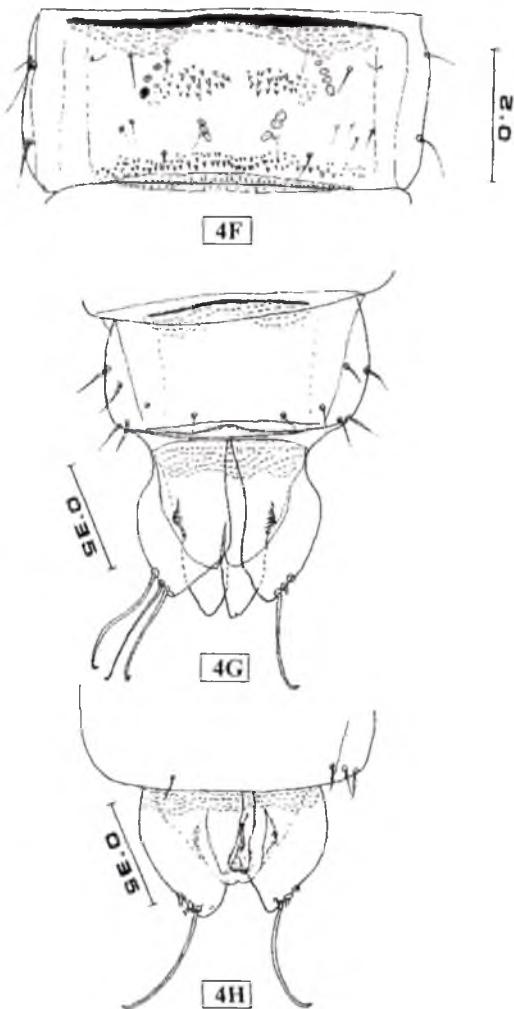


Fig. 4F–H: Pupa of *Orthocladius androgynus* Bhattacharyay, Ali and Chaudhuri. F. Tergite; G. Tergite VIII and IX (male) and H. Tergite IX (female)

claws (Fig. 3I). Anal tubules 4, narrow and bluntly tapered, 0.170 (0.140–0.200,  $n = 8$ ) long. Longest body seta 0.057 long.

*Pupa* Exuviae dark brown. Total body length 3.57 (3.55–3.61,  $n = 4$ ) in male and 3.69 in female. Length of thoracic horn/length of anal macroseta (Th/AM) 0.56.

*Cephalothorax*: Frontal seta 0.076 long, on frontal apotome (Fig. 4A); frontal warts absent. Antennal sheath (Fig. 4C) 0.930 long in male and 0.420 long in female. Anterior part of cephalothorax along eclosion line bumpy. Median antepronotals 0.136, 0.148, and lateral antepronotal 0.060 long. Thoracic horn (Fig. 4B) tubular, club-shaped, smooth, bare, 0.108 long and 0.030–0.032 wide. ThR 3.38. Anterior pre-

Table 2: Setae present on segment I–VIII

	I	II	III	IV	V	VI	VII	VIII
Dorsal (D)	3	5	5	5	5	5	5	2
Lateral (L)	1	3	3	3	3	3	4	4
Ventral (V)	2	4	4	4	4	4	4	1
O setae ( $O_d$ )	0	1	1	1	1	1	0	0

corneal seta 0.132, median seta 0.104 and posterior seta 0.116 long; of 4 dorsocentrals  $Dc_1$ ,  $Dc_2$  and  $Dc_3$  in a row;  $Dc_1$  0.048,  $Dc_2$  0.052,  $Dc_3$  0.080 and  $Dc_4$  0.020–0.024 long; distance between  $Dc_1$  and  $Dc_2$  0.172, between  $Dc_2$  and  $Dc_3$  0.024 and between  $Dc_3$  and  $Dc_4$  0.032. Wing sheath 1.05 long.

**Abdomen** (Fig. 4D–G): Tergite I without shagreen; T II with shagreen on anterior region with moderate bi median extension; T III–VIII with shagreen on anterior region with slight latero-median extension; T IX with shagreen present on anterior region (Fig. 4G–H). T III–VI with two well separated median clumps of strong posteriorly directed spines of which lateral spines are minute. T II–VII with transverse posterior rows of posteriorly directed spines; on tergites II–V the spines on both ends of the rows are clearly longer and stronger than those in the middle of the row forming a somewhat conspicuous patch. Conjectives II–V with transverse band of anteriorly directed sharply pointed fine spines lie posterior to the transverse row of posteriorly directed tergal spines. Sternites I–III without shagreen; IV–VIII with shagreen on anterior region; IX without shagreen. Pedes spurii B robust, present on segment II and III. Pedes spurii A well developed, only on segment VI consisting of broad patch of anteriorly directed spinules. Apophyses present on II–VIII, dark brown.

Anal lobe (Fig. 4G–H) rounded or oval, 0.270 (0.260–0.280,  $n = 4$ ) long and 0.270 wide; bearing 3 subequal apically hooked macrosetae, 0.186 (0.170–0.196,  $n = 4$ ) long. Genital sac in male 0.302 (0.284–0.232,  $n = 2$ ) and in female 0.170 long, in male overreaching anal lobe (Fig. 4G). G/F 1.13 (1.10–1.15,  $n = 2$ ) in male, and 0.74 (0.71–0.77,  $n = 2$ ) in female. ALR 1.92 (1.83–2.00,  $n = 2$ ) in male and 1.11 in female.

#### Material examined

2 larvae, West Bengal, Darjeeling (27° 03'N Lat., 88° 18'E Long., 2012 m msl), 01.V.92, Coll. D. K. Som; 1 larva, West Bengal, Darjeeling 20.V.92, Coll. D. K. Som; 1 male with larval and pupal exuviae (reared), 19.IV.92, Coll. D. K. Som; 1 male with larval and pupal exuviae (reared), 1.V.92, Coll. D. K. Som; *Holotype* male (Type No. B. U. Ent. 166) West Bengal, Mungpoo (26° 55'N Lat., 88° 28'E Long., Alt. 1149 m msl), 06.III.94, Coll. D. K. Guha; 2 females with larval and pupal exuviae (reared), 01.V.92, Coll. D. K. Som.

#### Remarks

Bhattacharya *et al.* (1991) first described the species in male from Darjeeling Himalayas of West Bengal. The reared males of the present study agree fully with *O. (E.) androgynus* Bhattacharya *et al.* and the females and immatures, not known beforehand are described here. The pupae are very similar to *O. (E.) mixtus* (Holmgren) in respect to the shagreen pattern on T II but differs in having no paired patches of spinules on T II of the former. The larva of the species has resemblance with *O. (E.)*.

*fuscimanus* (Kieffer) in having similar lauterborn organ and mandible. But the following combination of characters of imagines and immatures distinguish the species from other members of the genus *Orthocladius*:

**Adults:** i) AR in male 1.1–1.3 (1.29), in female 0.72, ii) scutellum with 10–12 setae, uniserial, iii)  $R_1$  with setae in both sexes, iv) squama with 12–14 setae, v) mid LR 0.45–0.47, vi) anal point pointed, bearing 3 setae on each side, vii) gonocoxite elongated and parallel shaped, viii) superior volsella elongated plate-like and distinct, ix) gonostylus short, x) female Gonocoxite IX with 2–3 setae and xi) seminal capsules dark snuff coloured.

**Pupa:** i) Thoracic horn club-shaped, smooth, ii) T II with anterior shagreen extending posteriorly on each side of the median line and without paired oral patches of stronger spinules, iii) segment VIII with 4 L setae of which 2 arranged on the posterior corner of the segment, iv)  $O_d$  setae present on segment II–VI, v) frontal setae present vii) presence of pedes spurii B on segment II and III and iii) pedes spurii A present only on sternite VI,

**Larva:** i) AR 1.79, ii) S I bifid, comparatively shorter, iii) head capsule 0.440 long, shorter in length, v) mentum 0.094 wide, vi) antennal blade very small, shorter than flagellum, 0.017 long and vii) anterior lacinial chaeta leaf-like. The subgenus *Eudactylo cladius* is represented relatively by a very specimens throughout the world and has undergone no revision (Langton and Cranston, 1991). The fascinating phenomenon observed in this species is frequent occurrence of spermatheca like structures in male adults and also in the body of some larvae. This may be due to infection of mermithid infection which was reported earlier (Wulker, 1961; Oliver, 1983). But interesting is that no noticeable morphological changes are observed in the infected species.

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of northeast India embodying the results of mosquito surveys carried out after 1973 in Assam and other north-eastern states. We have updated the systematic list of mosquito fauna recorded, so far, in Assam (Table 2) by incorporating the records of earlier studies on mosquito surveys carried out during pre and post DDT era with the present study which raises the total number of mosquito species to 101 in 14 genera and 25 sub genera from Assam. It includes genus *Aedeomyia* (1 sp.), *Aedes* (19 sp.), *Anopheles* (40 sp.), *Armigeres* (5 sp.), *Coquillettidia* (2 sp.), *Culex* (21 sp.), *Heizmannia* (1 sp.), *Mansonia* (4 sp.), *Malaya* (2 sp.), *Mimomyia* (1 sp.), *Orthopodomyia* (1 sp.), *Toxorhynchites* (1 sp.), *Tripteroides* (2 sp.) and *Uranotaenia* (1 sp.).

Table 3: Updated systematic list of mosquitoes ever recorded in Assam

Genus	<b><i>Aedeomyia</i></b> Theobald, 1901	
	Subgenus :	<b><i>Aedeomyia</i></b> Theobald, 1901
		– <i>catasticta</i> Knab, 1909
Genus	<b><i>Aedes</i></b> Meigen, 1818	
	Subgenus :	<b><i>Aedimorphus</i></b> Theobald, 1903
		– <i>caecus</i> (Theobald), 1901
		– <i>nigrostriatus</i> Barraud, 1927
		– <i>vexans</i> (Meigen), 1830
		– <i>vittatus</i> (Bigot), 1861
	Subgenus :	<b><i>Diceromyia</i></b> Theobald, 1911
		– <i>iyengari</i> Edwards, 1923
	Subgenus :	<b><i>Finlaya</i></b> Theobald, 1903
		– <i>albolateralis</i> (Theobald), 1908
		– <i>alboniveus</i> Barraud, 1934
		– <i>assamensis</i> (Theobald), 1908
		– <i>chrysolineatus</i> (Theobald), 1907
		– <i>dissimilis</i> (Leicester), 1908
		– <i>novoniveus</i> Barraud, 1934
		– <i>poeclius</i> Theobald, 1903
		– <i>pseudotaeniatus</i> Giles, 1901
	Subgenus :	<b><i>Mucidus</i></b> Theobald, 1901
		– <i>Scatophagooides</i> Theobald, 1901
	Subgenus :	<b><i>Neomelaniconian</i></b> Newstead, 1907
		– <i>lineatopennis</i> (Ludlow), 1905
	Subgenus :	<b><i>Stegomyia</i></b> Theobald, 1901
		– <i>aegypti</i> (Linnaeus), 1762
		– <i>albopictus</i> (Skuse), 1894
		– <i>annandalei</i> Theobald, 1910
		– <i>craggi</i> (?) Barraud, 1923
Genus	<b><i>Anopheles</i></b> Meigen, 1818	
	Subgenus :	<b><i>Anopheles</i></b> Meigen, 1818
		– <i>ahomi</i> Chowdhuri, 1929
		– <i>atkenii</i> James, 1903
		– <i>annandalei</i> Baini Prashad, 1918
		– <i>barbirostris</i> Van der Wulp, 1884
		– <i>bengalensis</i> Puri, 1930
		– <i>crawfordi</i> Reid, 1953
		– <i>gigas</i> Giles, 1901
		– <i>baileyi</i> Edwards, 1929
		– <i>insulaeflorum</i> (Swellengrebel and Swellengrewal de Graaf), 1919
		– <i>interruptus</i> Puri, 1929
		– <i>lindesayi</i> Giles, 1900

continued...

		— <i>nigerrimus</i> Giles, 1900 — <i>peditaeniatus</i> (Leicester), 1908 — <i>sinensis</i> Wiedemann, 1828 — <i>umbrosus</i> (Theobald), 1903
	Subgenus :	
		Cellia Theobald, 1902 — <i>aconitus</i> Doenitz, 1902 — <i>annularis</i> Van der Wulp, 1884 — <i>culicifacies</i> Giles, 1901 — <i>dirus</i> Peyton and Harrison, 1979 — <i>fluvialis</i> James, 1902 — <i>jamesii</i> Theobald, 1901 — <i>jeyporiensis</i> James, 1902 — <i>jeyporiensis</i> var. <i>candidiensis</i> Koidzumi, 1924 — <i>karwari</i> (James), 1903 — <i>kochi</i> Doenitz, 1901 — <i>maculatus</i> Theobald, 1901 — <i>majidi</i> Young and Majid, 1928 — <i>minimus</i> Theobald, 1901 — <i>nivipes</i> (Theobald), 1903 — <i>pallidus</i> Theobald, 1901 — <i>philippinensis</i> Ludlow, 1902 — <i>pseudojamesii</i> Strickland and Chowdhury, 1931 — <i>splendidus</i> Koidzumi, 1920 — <i>stephensi</i> Liston, 1901 — <i>subpictus</i> Grassi, 1899 — <i>tesselatus</i> Theobald, 1901 — <i>theobaldi</i> Giles, 1901 — <i>vagus</i> Doenitz, 1902 — <i>varuna</i> Iyenger, 1924 — <i>willmorei</i> (James), 1903
Genus	<b>Armigeres</b> Theobald, 1901	
	Subgenus :	
		— <i>kuchingensis</i> Edwards, 1915 — <i>subalbatus</i> (Coquillett), 1898
	Subgenus :	
Genus	<b>Coquillettidia</b> Dyar, 1905	
	Subgenus :	
Genus	<b>Culex</b> Linnaeus, 1758	
	Subgenus :	
		— <i>bitaeniorhynchus</i> Giles, 1901 — <i>cornutus</i> Edwards, 1922 — <i>epidesmus</i> (Theobald), 1910 — <i>fuscocephala</i> Theobald, 1907 — <i>gelidus</i> Theobald, 1901 — <i>mimeticus</i> Noe, 1899 — <i>mimulus</i> Edwards, 1915 — <i>pseudovishnui</i> Colless, 1957 — <i>quinquefasciatus</i> Say, 1823 — <i>sinensis</i> Theobald, 1903

continued ...

			– <i>vishnui</i> Theobald, 1901
			– <i>whitmorei</i> (Giles), 1904
		<b>Culiciomyia</b> Theobald, 1907	
		– <i>bailyi</i> Barraud, 1934	
		– <i>pallidothorax</i> Theobald, 1905	
		<b>Eumelanomyia</b> Theobald, 1909	
		– <i>brevipalpis</i> (Giles), 1902	
		– <i>malayi</i> (Leicester), 1908	
		<b>Lophoceraomyia</b> Theobald, 1905	
		– <i>minor</i> (Leicester), 1908	
		– <i>peytoni</i> Brans and Rattnarathikul, 1967	
		<b>Lutzia</b> Theobald, 1903	
		– <i>fuscanus</i> Wiedemann, 1820	
		– <i>halifaxii</i> Theobald, 1903	
<b>Genus</b>	<b>Heizmannia</b> Ludlow, 1905		
	<b>Subgenus :</b>	<b>Heizmannia</b> Ludlow, 1905	
<b>Genus</b>	<b>Malaya</b> Leicester, 1908	– <i>reidi</i> Mattingly, 1957	
	<b>Subgenus :</b>	<b>Maorigoeldia</b> Edwards, 1930	
<b>Genus</b>	<b>Mansonia</b> Blanchard, 1901	– <i>genurostris</i> Leicester, 1908	
	<b>Subgenus :</b>	– <i>jacobsoni</i> Edwards, 1930	
<b>Genus</b>	<b>Mimomyia</b> Theobald, 1903	<b>Mansonioides</b> Theobald, 1907	
	<b>Subgenus :</b>	– <i>annulifera</i> (Theobald), 1901	
<b>Genus</b>	<b>Orthopodomyia</b> Theobald, 1904	– <i>dives</i> (Schiner), 1968	
<b>Genus</b>	<b>Toxorhynchites</b> Theobald, 1901	– <i>indiana</i> Edwards, 1930	
	<b>Subgenus :</b>	– <i>uniformis</i> (Theobald), 1901	
<b>Genus</b>	<b>Tripterooides</b> Giles, 1904	<b>Mimomyia</b> Theobald, 1903	
	<b>Subgenus :</b>	– <i>chamberlaini</i> Ludlow, 1904	
	<b>Subgenus :</b>	– <i>anopheloides</i> (Giles), 1903	
<b>Genus</b>	<b>Uranotaenia</b> Lynch Attribalzaga, 1891	<b>Toxorhynchites</b> Theobald, 1901	
		– <i>splendens</i> (Wiedemann), 1819	
		<b>Rachionotomyia</b> Theobald, 1905	
		– <i>aranoides</i> (Theobald), 1901	
		<b>Tripterooides</b> Giles, 1904	
		– <i>indicus</i> (Barraud), 1929	
		– unidentified	

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# **Mosquito Fauna in a Broken Forest Ecosystem of District Dibrugarh with Updated Systematic List of Mosquitoes Recorded in Assam, India**

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**Abstract:** Mosquito surveys in a broken, inhabited forest fringed area of Dibrugarh district, Assam yielded 50 species of mosquitoes in 11 genera of which *Culex peytoni*, *Culex bailyi* and *Heizmannia reidi* were the new records from Assam. An updated systematic list of mosquitoes ever recorded in Assam has been provided.

**Keywords:** Check list, Mosquito survey, New record

## **INTRODUCTION**

Mosquito surveys provide valuable information on occurrence, distribution, prevalence and species composition of various mosquitoes in an area which assumes significance due to their public health importance. The state of Assam, situated in the north-eastern region of India between 24.13°–28.0° N and 89.46°–96.40° E, is rich in diversity of flora and fauna due to its typical climatic conditions and geographical features. In the pre DDT era, Christophers (1933) and Barraud (1934) published their monumental monographs on anopheline and culicine fauna of British India respectively which included mosquitoes of Assam also. The first ever anopheline survey in Assam was carried out by Bentley (1902) in Tejpur area. Later surveys were conducted by Chal-lam (1923) in Kamrup district, Strickland (1929) and Ramsay (1930) in Cachar, Gupta and Majumdar (1932) in Goalpara, Rice and Savage (1932) in upper Assam areas, Viswanathan *et al.* (1941) in various parts of Assam. However, all these surveys were aimed at anopheline fauna as these studies were related with malaria. In post DDT era mosquito fauna surveys were carried out during 1966–67 jointly by Zoological Survey of India and Defence Research laboratory, Tejpur in Assam and NEFA. Other important mosquito surveys during last 2 decades in different parts of Assam were conducted by

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Sarkar *et al.* (1981, 1984), Karim *et al.* (1985), Nagpal and Sharma (1987) and Anil Prakash *et al.* (1997). In order to add to the existing information on mosquitoes of Assam, we carried out an intensive fauna study in a broken, inhabited forest area of Dibrugarh district the results of which are presented in this communication. In addition, an updated systematic list of mosquitoes recorded in Assam, so far, has been compiled after incorporating the present results.

#### MATERIAL AND METHODS

The year long study was carried out during 1995–96 in a forest fringed village viz. Soraipung belonging to Tengarhat Primary Health Centre, District Dibrugarh, Assam. The village is situated at the distance of about 100 meters from the fringes of a broken, evergreen, tropical forest area viz. 'Soraipung forest range'. Peridomestic ditches, kuccha wells, pits, pools, paddy fields, streams in the inhabited village area and ground pools, elephant foot prints, marshy areas, streams and tree holes in the adjoining uninhabited forest area were the main breeding habitats of mosquitoes in the study area.

Both immature's and adult mosquito collections were carried out at monthly intervals between August 1995 and July 1996 from the study area (village and the adjoining forest). Adult mosquitoes were collected from dusk to dawn in the village (i) on indoor human bait and (ii) with CDC light traps from human dwellings and cattle sheds. In addition, day resting adult collections were carried out indoors in village human dwellings and outdoors in forest areas. Mosquito immatures were collected from different breeding habitats, available in the village and forest areas, and link reared to adult stage individually in plastic vials. Mosquitoes were identified using the keys of Christophers (1933), Barraud (1934), Reid (1968), Mattingly (1970) and Sirivanakarn (1977).

#### RESULTS AND DISCUSSION

Mosquitoes belonging to 11 genera and 50 species were collected in the present study (Table 1). Collections by different methods yielded adult mosquitoes of 7 genera and 34 species, whereas, immatures of 34 species in 8 genera were captured. Breeding habitats of mosquitoes collected as immatures are listed in Table 2.

In adult collections both *Anopheles philippinensis* and *Anopheles nivipes* were recorded in 60 : 40 ratio. These 2 species are closely related and differentiated by basal extension of presector dark mark on wing vein 1 beyond the distal end of humeral dark mark on costa in *Anopheles nivipes* adult females. However, on link rearing all the emerged adults from the immatures were identified as *Anopheles nivipes* on the basis of larval and pupal characters as described by Reid (1968). It indicates that identification of these two species solely on wing character will be erroneous and that examination of larval and pupal characters are necessary to separate these species. The study of Nagpal and Sharma (1987) on mosquito fauna of north-eastern states also demonstrated that prevalent species understood to be resembling the identification of *Anopheles philippinensis* was in fact *Anopheles nivipes*. Further studies are required to know the exact status of these two species in the north-eastern region.

Eight species of mosquitoes in 4 genera were collected breeding in tree holes from the forest area (Table 2). These are *Culex peytoni*, *Tripteroides indicus*, *Heizmannia*

Table 1: Mosquito collected from Soraipung area, Dibrugarh, Assam

Sl. No	Genus	Subgenus	Total Sp.	Species name(s)
1.	Aedeomyia	Aedeomyia	1	catasticta
2.	Aedes	Aedimorphus	4	caecus, nigrostriatus, vexans, vittatus (?)
		Finlaya	3	albolateralis, assamensis, chrysolineatus
		Neomelaniconion	1	lineatopennis
		Stegomyia	2	aegypti, craggi / annandalei (?)
3.	Anopheles	Anopheles	4	barbirostris, baileyi, nigerrimus, peditaeniatus
		Cellia	11	aconitus, annularis, dirus, kochi, maculatus, nivipes, philippinensis, splendidus, tesselatus, vagus, varuna
4.	Armigeres	Armigeres	1	subalbatus
5.	Coquillettidia	Coquillettidia	1	unidentified
6.	Culex	Culex	9	bitaeniorhynchus, fuscocephala, gelidus, mimulus, pseudovishnui, quinquefasciatus, tritaeniorhynchus, vishnui, whitmorei
		Culiciomyia	2	bailyi*, pallidothorax
		Lophoceraomyia	1	peytoni*
		Lutzia	2	fuscanus, halifaxii
7.	Heizmannia	Heizmannia	1	reidi*
8.	Mansonia	Mansonioides	4	annulifera, dives, indiana, uniformis
9.	Orthopodomyia	—	1	anopheloides
10.	Tripteroides	Tripteroides	1	indicus
11.	Uranotaenia	Unidentified	1	unidentified

\* New record

*reidi*, *Aedes albolateralis*, *Aedes assamensis*, *Aedes chrysolineatus*, *Aedes vittatus* (?), and *Aedes craggi/ annendelai* (?). *Aedes craggi* and *Aedes annendelai* are closely related species, separable only on the basis of male genitalia. The species confirmation between *craggi* and *annendelai* could not be done due to the non emergence of male mosquito from the immatures. Similar was the case with *Aedes vittatus*. Recently Bhattacharyya *et al.* (1995) described the larva and pupa of *Aedes nigrostriatus* from Dibrugarh district. This mosquito, adult of which were first described by Barraud in 1927 from Golaghat area of Assam (Barraud, 1934), was collected in good numbers in the present study in the ground pools in village as well as forest areas.

#### NEW RECORDS

Based on the published literature, *Culex peytoni*, *Culex bailyi* and *Heizmannia reidi* in the present study were collected for the first time from Assam, thus, constituting new records. *Culex peytoni* though has been recorded from Andamans (Sirivanakarn, 1977), ours is the first record of this mosquito from mainland India (Bhattacharyya *et al.*, 1998). Similarly, *Heizmannia reidi*, recorded from Sukna, Darjeeling District, West Bengal, India (Mattingly, 1970), has been collected for the first time from Assam by us (Bhattacharyya *et al.*, 1998). *Culex bailyi*, known only from its type locality i.e. Virajpet (Coorg) in south India (Barraud, 1934), is also the first record from Assam. This species was collected in good numbers from shaded pools and elephant foot prints

Table 2: Breeding habitats of mosquitoes collected as immatures from Soraipung area, Dibrugarh, Assam.

Species	Breeding habitats in village area						Breeding habitats in forest area				
	Ditch	Kuc-cha	Drain	Gro-und	Stream	Paddy	Gro-und	Animal	Mar-shy	Stream	Tree
	well	well	pool	pool	bed	field	pool	foot	area	bed	hole
<i>Aedeomyia catasticta</i>	x	—	—	—	—	—	—	—	—	—	—
<i>Aedes albolateralis</i>	—	—	—	—	—	—	—	—	—	—	x
<i>Ae. assamensis</i>	—	—	—	—	—	—	—	—	—	—	x
<i>Ae. caecus</i>	x	—	—	—	—	—	x	x	—	—	—
<i>Ae. chrysolineatus</i>	—	—	—	—	—	—	—	—	—	—	x
<i>Ae. craggi/annandalei</i>	—	—	—	—	—	—	—	—	—	—	x
<i>Ae. nigrostriatus</i>	—	—	—	—	—	x	x	x	—	—	—
<i>Ae. vittatus</i>	—	—	—	—	—	—	—	—	—	—	x
<i>Anopheles aconitus</i>	x	—	—	—	—	—	x	—	—	—	—
<i>An. baileyi</i>	—	—	—	—	—	—	x	x	—	—	—
<i>An. barbirostris</i>	x	x	—	x	x	—	x	—	x	—	—
<i>An. dirus</i>	—	—	—	—	—	—	x	x	—	x	—
<i>An. kochi</i>	x	x	—	—	—	x	x	x	—	—	—
<i>An. maculatus</i>	—	—	—	—	—	—	x	—	—	—	—
<i>An. nivipes</i>	x	—	—	—	—	—	x	—	—	—	—
<i>An. peditaeniatus</i>	x	x	x	x	x	x	x	—	x	x	—
<i>An. splendidus</i>	x	—	—	—	—	—	—	—	—	—	—
<i>An. vagus</i>	x	x	—	x	—	—	x	x	—	—	—
<i>An. varuna</i>	x	—	—	—	—	—	—	—	—	—	—
<i>Culex bailyi</i>	x	x	—	x	—	—	x	x	—	x	—
<i>Cx. bitaeniorhynchus</i>	—	x	x	—	x	—	—	—	—	—	—
<i>Cx. fusciceps</i>	x	x	—	—	—	—	x	—	—	—	—
<i>Cx. helifaxii</i>	x	—	x	—	—	—	x	x	—	—	—
<i>Cx. mimulus</i>	—	—	—	—	—	—	x	—	—	—	—
<i>Cx. pallidothorax</i>	—	—	—	—	—	—	x	x	—	—	—
<i>Cx. peytoni</i>	—	—	—	—	—	—	—	—	—	—	x
<i>Cx. pseudovishnui</i>	x	—	x	—	x	—	—	—	—	—	—
<i>Cx. quinquefasciatus</i>	—	—	—	—	—	—	x	—	—	—	—
<i>Cx. tritaeniorhynchus</i>	x	x	—	x	—	—	—	—	—	—	—
<i>Cx. vishnui</i>	x	x	x	x	—	—	—	—	—	—	—
<i>Heizmannia reidi</i>	—	—	—	—	—	—	—	—	—	—	x
<i>Orthopodomyia anopheloides</i>	x	—	—	—	x	—	x	—	—	—	—
<i>Tripteroides indicus</i>	—	—	—	—	—	—	—	—	—	—	x
<i>Uranotaenia sp.</i>	—	—	—	—	—	—	x	—	—	—	—

x = Species Present

in forest areas in association with *Anopheles baileyi* and *Culex pallidothorax* in January 1996. In May 1996 also, 1 larva of *Culex bailyi* was collected from a peridomestic ditch in the village area. This species was separated from its closely related species i.e. *Culex pallidothorax* on the basis of male genitalia following Barraud (1934).

Sarkar *et al.* (1981) recorded 27 species of mosquitoes in 8 genera from Dibrugarh district of Assam. Later, Nagpal and Sharma (1987) reported 59 species of mosquitoes under 8 genera from Assam. Recently Malhotra and Mahanta (1994) listed 79 mosquito species in 10 genera and 21 sub genera from Assam in their checklist of mosquitoes

# Population Dynamics and Host Specificity of Flea Beetle *Altica himensis* Shukla (Coleoptera: Chrysomelidae: Alticinae) in Kumaon Himalayas

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**Abstract:** Ecological studies on flea beetles were conducted in a Kumaon Himalayan Oak mixed forest for two years, 1994 and 1995. The beetle population was found confined to herb strata. Six host plants are reported.

Interspecific and Intraspecific host discrimination was shown and considerable damage caused to host plants. The seasonal fluctuations were observed in the population of adult and juveniles over an annual cycle. Density of adults varied from  $1.5/m^2$  (March)- $18.0/m^2$  (May); eggs from  $40.50/m^2$  (November)- $250.0/m^2$  (July). Larval density fluctuated from  $12.50/m^2$  (November)- $70.55/m^2$  (August). It is a multivoltine insect, three peaks of overlapping generations in May, July and September were observed. Population was found to be host specific. From November to May it remained confined to *R. hastatus* whereas, with the onset of rainy season got shifted to other host plants (mostly summer annuals) and flourished on them till early winter (October–November).

**Keywords:** Phytosociology, Population dynamics, Flea-beetle, Weed, Host preference and Biological control.

## INTRODUCTION

*Altica* species are distributed throughout the world. These are phytophagous attacking crops, vegetables, fruits, ornamentals and forest trees (Picard, 1913; Port and Guile, 1986; Mineo and Iannazzo, 1986; Cabral and Ferreira, 1991; Kapoor, 1993). Some species are reported to possess the potential for biological control of weeds (Zwölfer, 1965; Goeden, 1983; Ooi, 1987; Nayek and Bañerjee, 1987; Hill, 1989; Napompeth, 1991). Alticinae beetles are generally specific in their food plant choice (Biondi, 1990; Joseph, 1991). A few studies have been made on population dynamics of flea beetles (Ooi, 1987; Jhala *et al.*, 1987; Michaud, 1990).

Screening of literature reveals that no work has been done on *Altica* species in the Kumaon Himalayas. Thus the present work attempts to study population fluctuations and host specificity of flea beetles in the region.

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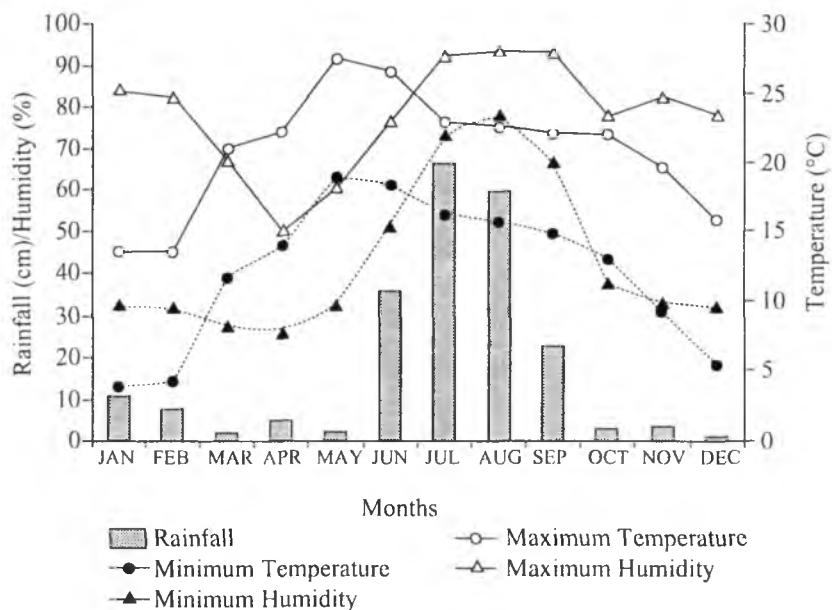


Fig. 1: Ombrothermic diagram for Nainital (1994–1995)

## MATERIALS AND METHODS

The study site, a mixed oak forest was selected near D. S. B. Campus, Naini Tal (29°23'N and 79°28'E, altitude 2050 m). Weather data was obtained from the nearest station, State Observatory, Naini Tal (Fig. 1). On the basis of climatic variations, the year is divisible into three seasons, summer (March–June), rainy (July–October) and winter (November–February). Random sampling with the help of 1 m × 1 m quadrat was done and phytosociological parameters, *viz.*, frequency, abundance, density and distribution-ratio were calculated for host plants, following Curtis and Cottam (1956) and Misra (1968), as follows:

$$\begin{aligned}
 \text{Frequency (\%)} &= \frac{\text{No. of sampling units in which the species occurred}}{\text{Total no. of sampling units studied}} \times 100 \\
 \text{Abundance} &= \frac{\text{Total no. of individuals of the species in all the sampling units}}{\text{No. of sampling units in which the species occurred}} \\
 \text{Distribution ratio} &= \frac{\text{Abundance}}{\text{Frequency}} \\
 \text{Density} &= \frac{\text{Total no. of individuals of the species in all the sampling units}}{\text{Total no. of sampling units studied}}
 \end{aligned}$$

The adults, juveniles and eggs of flea beetles were also sampled by direct counting per quadrat of 1 m × 1 m (Southwood, 1978). Quadrat sampling was preferred over per plant sampling because the beetle population was found to occur in a variety of

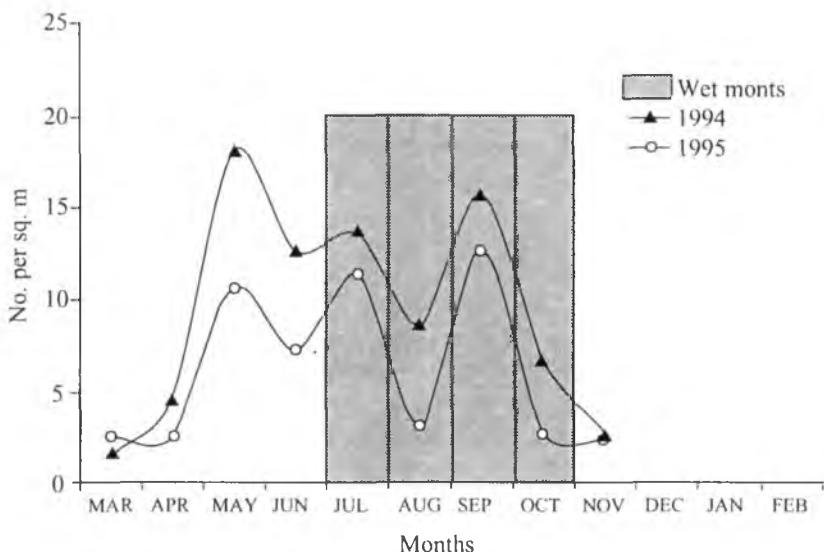


Fig. 2: Seasonal variation in population density of adult *A. himemesis*

host plants of varying characteristics. A hinged tray with white painted surfaces and raised rims was placed under the plants, enclosing the base of the plant completely (Croix, 1961). Then the plants were tapped gently. Beetles quickly fell from their host plant into the tray and were counted immediately before they could hop off the tray. Extreme care was taken while approaching the quadrat/tray, as flea beetles hop out very quickly. Ten quadrats were sampled every fortnight and calculated average was plotted against month of occurrence. A preliminary "no choice test" on host specificity was performed following Nayek and Banerjee (1987). Larvae and adults were exposed to a series of plant species of similar age, under laboratory condition. A series of potted plants were taken. Each pot measured  $20\text{ cm}^2 \times 15\text{ cm}$  high and was  $2/3^{\text{rd}}$  filled with sterilized soil. Three young plants of each test species, 8–9 cm in height, were grown in pots. Pots were covered by a nylon-net-cage ( $20\text{ cm}^2 \times 30\text{ cm}$  high, fine mesh). Cages fit tightly on to rims of pots. Twenty newly emerged paired adults or 30 mixed sex and aged larvae were put into each cage. Plants were regularly watered. After three days, plants were examined daily for feeding and oviposition. Feeding was determined on a subjective visual scale. Tests were terminated after ten and fifteen days of larval and adult exposure, respectively. A pot containing plant but no insect was also kept as a "Control". Tests were repeated three times.

After this experiment, a "choice test" was performed. Adults and larvae were offered a choice between *Impatiens amplexicaulis* and one of the other five plant species (*Impatiens scabrida* DC and *I. racemosa* DC (F. Balsaminaceae); *Rumex hastatus* D Don and *R. nepalensis* Spreng (F. Polygonaceae) and *Oenothera rosea* G Don (F. Onagraceae)). Two plants of each test species were grown together in caged pots and the same process was repeated as in "no choice test".

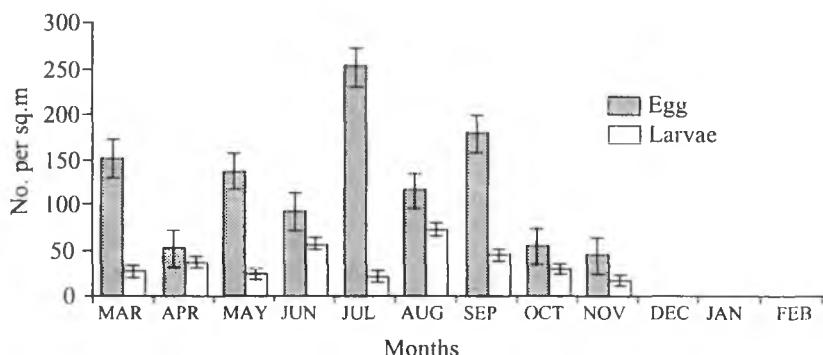


Fig. 3: Seasonal variation in eggs and larval density of *A. himensis*

## RESULTS AND DISCUSSION

The minimum temperature and humidity varied 2.8°C–21.2°C and 20.9%–78.3%, respectively with annual rainfall being 2233 mm during the study periods. Phytosociology of six host plants revealed that distribution ratio ranged from 0.27 to 0.80, indicating contagious or patchy distribution of host plants. Frequency varied from 5% (*I. racemosa*) to 15% (*I. amphorata*) and density ranged between 1.10 tiller /m<sup>2</sup> (*I. racemosa*) and 3.24 tiller /m<sup>2</sup> (*R. hastatus*). Host plants of *A. himensis* were confined to herb strata.

Population estimation showed marked seasonal variations in the density of adults as well as juveniles (Figs. 2 and 3). Density of adults varied from 1.5 /m<sup>2</sup> (March) to 18.0 /m<sup>2</sup> (May) in 1994 and from 2.2 /m<sup>2</sup> (November) to 12.5 /m<sup>2</sup> (September) in 1995. Egg-density ranged between 40.50 /m<sup>2</sup> (November) and 250.0 /m<sup>2</sup> (July), whereas larval density fluctuated from 12.50 /m<sup>2</sup> (November) to 70.53 /m<sup>2</sup> (August). Cumulative values of 1994 and 1995 for larval and egg density) are shown in Fig. 3.

Two year study has recorded maximum density of adults during or towards the end of the wet-period, 13.5 /m<sup>2</sup> (July)-15.5 /m<sup>2</sup> (September) followed by summer period, 1.5 /m<sup>2</sup> (March)-12.5 /m<sup>2</sup> June and early winter, 2.5 /m<sup>2</sup> (November)-6.5 /m<sup>2</sup> (October). Lower values obtained in 1995 can be attributed to the lower primary productivity in this year. Density of *I. amphorata*, the most preferred host was lesser in 1995 (personal observation). Same trend was followed by juveniles. Three distinct peaks of population density were recorded i.e. in May, July and September. Environmental stress (parasites, predators and climate) was greatly felt by the population at each stage of development. From December to February the population of *Altica himensis* is found to hibernate as adult under dry leaves and debris near *R. hastatus* plants.

Population density of adult coleopterans has been studied by various workers. Low temperature and low humidity in winter as well as high temperature and low humidity during summer is found responsible for lower population estimates during these months (Dempster, 1975; Kaushal and Vats, 1987). Low primary productivity during these periods is also found responsible for lower estimates (Clark *et al.*, 1967).

Table 1: Plants tested for feeding by *A. himensis* larvae and adults in the laboratory

Name of Host plant	Adult feeding			Oviposition site	Larval feeding			
	Leaf	Petiole	Stem		Leaf	Petiole	Stem	
<i>Impatiens amphorata</i> Edgew.	+	±	±*	±*	Lower leaf surface	+	±*	±*
<i>I. racemosa</i> DC.	+	±	—	—	Lower leaf surface	+	—	—
<i>I. scabrida</i> DC.	+	—	—	—	Lower leaf surface	+	—	—
<i>Oenothera missa</i> D. Don.	+	—	—	—	Lower leaf surface	+	—	—
<i>Rumex hastatus</i> D. Don.	+	—	—	under the sheath of petiole	Lower leaf surface & petiole	+	±*	±*
<i>Rumex nepalensis</i> Sprengel Seasonal ornamental plant /others*	—	—	—	—	—	±	—	—
	**	—	—	—	—	**	—	—

\* - Feeding; ± - Slight feeding; - - No feeding; \* - Feeding by third instar larva and adults occurred only in the absence of leaves; \*\* - Insects starved

Jhala *et al.* (1987) reported average adult density of *A. cyanea*, on weeds in paddy fields. Density was recorded as, 17.40, 19.0 and 54.40 adults per plant of *Ammania baccifera*, *Ludwigia parviflora* and *Spermacoce hispida*, respectively. Ooi (1987) recorded an average of  $1.3 \pm 0.4$  adults and  $7.7 \pm 2.0$  larvae per plant of *Melastoma malabathricum*. Michaud (1990) reported egg density of  $10.0/m^2$ ,  $200/m^2$  and  $400/m^2$  at 185 m, 615 m and 830 m elevations, respectively, on weed *Epilobium angustifolium*. Lal (1977) reported *Altica caeruleascens* overwintering as adult under the bark of willow (*Salix alba*).

Results of "no choice test" on host specificity are presented in Table 1. In absence of leaves adults chewed the petioles and stems of *I. amforata*, whereas floral feeding was done by larvae on *I. amforata*, *I. scabrida* and *R. hastatus*.

Host preference order is observed as follows:

*I. amforata* > *I. scabrida* > *I. racemosa* > *R. hastatus* > *O. rosea* > *R. nepalensis*.

Polyphagy as shown by the beetle is attributed to availability of host plants presented with young leaves during rainy season (Thomas, 1987). It is interesting to note that the population of *A. himensis* is supported throughout the winter (November–February) and early summer (March–May) by dock-weed, *R. hastatus* only. Other host plants appear with the onset of rains and bear young delicate leaves, whereas, *R. hastatus* leaves have grown mature, sclerotized and stem grew woody by this period. By the end of October, all summer annuals perished and population finally shifted to *R. hastatus*, the reservoir host for winter refuge.

Considerable damage is caused to *Impatiens* species complex (85%). Their abundance affected markedly as these plants propagate by seed formation and beetle population reduced their vitality to great extent.

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## Multicellular tray for rearing the larvae of *Helicoverpa armigera* (Hübner) (Lepidoptera: Noctuidae)

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**Abstract:** A multicellular larval rearing unit has been devised for rearing *Helicoverpa armigera* (Hübner). The tray is made of transparent acrylic and is amenable to surface sterilisation. It is reusable, durable and hence economical. Rearing in this tray provided 80 to 90% recovery of *H. armigera* pupae. Larval escape was nil and disease incidence by contamination was minimum. Cannibalism was totally avoided. Pupal formation could be recorded easily and pupal collection was also easy.

**Keywords:** *Helicoverpa armigera*, multicellular tray, cannibalism, pupation

### INTRODUCTION

A major problem encountered by research workers in culturing both *Heliothis* and *Helicoverpa* spp. has been that of cannibalism (Hardwick, 1965; Twine, 1971; Whitlock, 1973; Odindo, 1981). To overcome this problem, most of the workers have recommended rearing larvae in isolation (Griffith and Smith, 1977; Teakle and Jensen, 1985). But these isolated rearing techniques are time consuming and they do not permit the inherent locomotory behaviour of the larvae to occur owing to inadequate space in the container. This is also considered to be contrary to the natural ecology of the larvae (Griffith and Haskell, 1988). Hence, the other system which is in use for large scale production of *Heliothis* spp. is the use of compartmentalised trays, which have been used earlier for rearing corn earworm, *Heliothis zea* (Boddie) (Sparks and Harrel, 1976) and *Heliothis virescens* (F.) (Raulston and Lingren, 1972). The major problem associated with rearing of *Helicoverpa armigera* (Hübner) (Lepidoptera: Noctuidae) is also the high mortality caused by cannibalism and disease incidence. The present study was taken up with an intention to alleviate the above mentioned problems and to develop a rearing unit for multiplying *H. armigera* in large numbers in research laboratories and in commercial insectaries.

## MATERIALS AND METHODS

The newly developed multicellular larval rearing unit was compared with two other types of larval rearing units for the rearing of *Helicoverpa armigera*. The semi-synthetic diet utilised in this study was the one developed by Nagarkatti and Sathyaprakash (1974). The eggs of *H. armigera* were collected from the mass production unit of the Project Directorate of Biological Control, Bangalore, India. The eggs were collected on a piece of cloth after surface sterilisation (with 0.1% sodium hypochlorite solution) and fixed to the underside of the lid of a ventilated bread box (measuring 21 × 11 × 6 cm) containing 400 ml of semi-synthetic diet. The larvae, on hatching, moved on to the diet and started feeding. When the larvae were 4 to 5-day-old (2nd instar), they were transferred to three different larval rearing units - diet bottles, plastic louvers and the newly devised multicellular larval rearing unit. The efficiency of the three units was compared.

### Diet bottle

(4 cm height, 2 cm diameter, each could hold 8 gm of diet and amenable to heat sterilisation). The semi-synthetic diet was prepared and 1100 ml of diet was immediately poured into about 135 vials. After the diet solidified and cooled, 4 to 5-day-old larvae were transferred individually into each diet bottle and plugged with cotton.

### Plastic louver

(measuring 60 × 22 cm with 200 cells, each cell measured 2.5 × 2.5 × 1.5 cm and could hold 5 gm of diet with 2 acrylic sheets (62 × 24 cm)). This unit was amenable to surface sterilisation (initially washed in hot water and soaked in 1% sodium hypochlorite solution for one hour and further rinsed in distilled water). The louver was placed on one acrylic sheet, semi-synthetic diet was prepared and 1100 ml was poured into the cells. After the diet cooled and solidified, 4 to 5-day-old larvae were transferred into the louver @ one larva per cell and then covered the louver with the other acrylic sheet (which was provided with minute holes for aeration). The complete set up was fastened with metal clips along the edges.

### Multicellular larval rearing unit

This tray is a modification of the larval rearing unit described by Singh and Rembold (1992). The modifications were made to suit the existing conditions of rearing, using readily available materials. The cost of the tray was approximately Rs. 1400. This unit was made up of two rectangular trays and a lid lined with wire mesh (all made of 4 mm thick acrylic sheet) (Fig. 1A). The outer or basal tray measured 37 × 32 × 5 cm and the inner or upper tray 36 × 31 × 5.5 cm and these two were completely separable. The inner tray was divided into 100 compartments, each compartment measured 3.5 × 3 × 4.5 cm and could hold 11 gm of diet. The inner tray could exactly fit into the inner part of the outer tray. In the central floor of each compartment, a small circular hole was provided (1.4 cm diameter). This unit was amenable to surface sterilisation (initially washed in hot water and soaked in 1% sodium hypochlorite solution for one hour and then rinsed in distilled water). The semi-synthetic diet was prepared and 1100 ml of diet was poured into the basal tray. After the diet solidified and cooled, the inner tray was inverted and placed over the outer tray, with the holes facing upwards

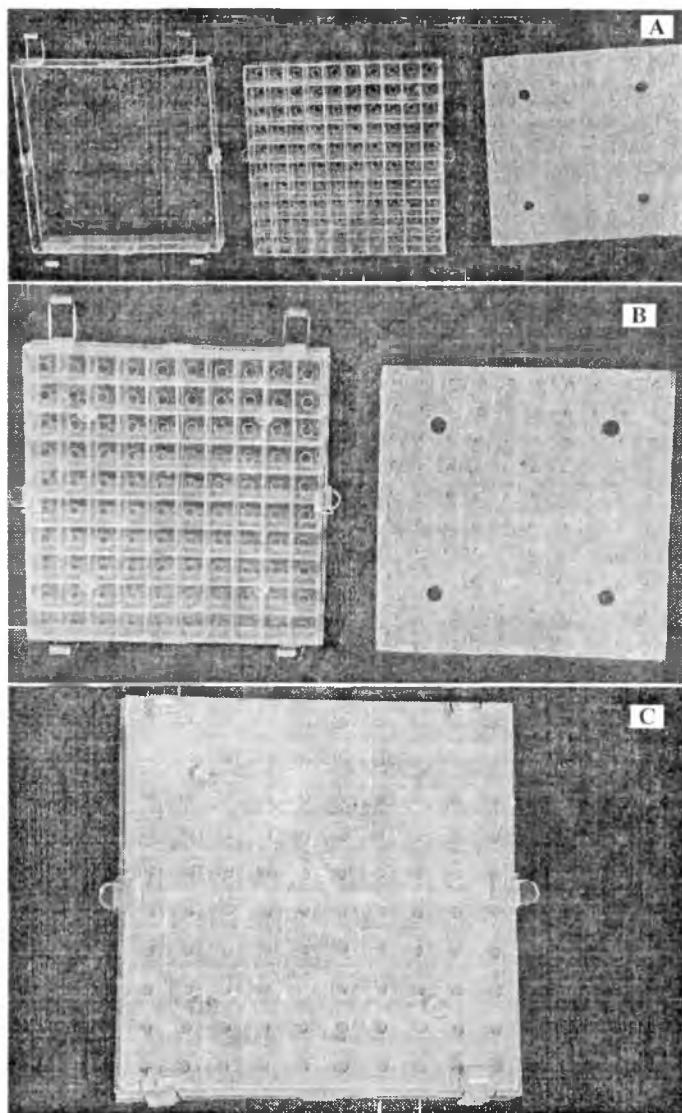


Fig. 1: Multicellular larval rearing unit (A) comprising two trays and a lid (B) multicellular tray inverted over the plain tray (C) complete set up of the multicellular larval rearing unit

(Fig. 1B). The compartments in the upper (inner) tray divided the diet in the basal tray into pieces. The larvae (4 to 5-day-old) could be individually transferred through the hole into each compartment. The lid (with holes exactly corresponding to the holes on the inner tray) lined with wire mesh was then used to cover the structure (Fig. 1C). Then the clamps were fixed. The basal tray had handles on both sides used for carrying the trays. Knobs were provided at the base of the inner tray which enabled the stacking

Table 1: Comparative efficiency of three kinds of larval rearing units

Parameters	Larval rearing units			CD at $P \leq 0.01$
	Diet bottles	Plastic louver	Multicellular tray	
Per cent pupation	75%	28%	83%	18.3
Per cent adult emergence	90%	75%	88%	NS
Per cent larvae escaped/mortality due to cannibalism	0%	35.1%	1.2%	15.3%
Per cent larvae diseased	10.2%	30.3%	1.5%	12.2
Per cent mortality due to unknown reasons	4.75%	6.9%	9.1%	NS
Weight of pupa	356.2 mg	340.4 mg	367.8 mg	NS
Cost of rearing 100 pupae	Rs. 156.00	Rs. 424.00	Rs. 118.00	*
Capacity of each cell/bottle (gm of diet)	8 gm	5 gm	11 gm	*
Time required for (a) dispensing of diet and transfer of larvae	30 mins	30 mins	15 mins	*
(b) collection of 100 pupae	35 mins	15 mins	15 mins	*

Note: \*Not subjected to statistical analysis

of trays.

All the three units were placed in BODs set at  $26 \pm 2^\circ\text{C}$  and  $65 \pm 5\%$  RH. The performance of each type of larval rearing unit was evaluated based on twenty replications. The data obtained on - per cent pupation, per cent adult emergence, per cent larvae which escaped or moved to neighbouring cells, per cent larvae diseased and weight of pupae - were subjected to ANOVA. The cost involved in rearing and time involved in dispensing the diet, larval transfer and pupal collection in the three larval rearing units were also compared. For calculating the cost of rearing, the diet cost, the life of the rearing unit, the amount of labour involved, the per cent pupation, etc. were taken into consideration.

## RESULTS AND DISCUSSION

### Individual rearing in diet bottles

The major advantages in using the diet bottles were: (a) 60-80% survival was obtained (b) the pupal weight ranged from 320 to 389 mg (c) the bottles could be heat-sterilised and (d) cannibalism and escape of larvae and spread of diseases were minimum. The disadvantages were: (a) each diet bottle could hold only 8 gm of diet and it was not sufficient for the total development of the larvae (b) it was necessary to further transfer each growing larva into another vial with about 3 to 4 gm of diet for completing the larval development, hence care had to be taken not to dispense excess diet in the second set of diet bottles, which could lead to wastage of diet (c) disease incidence was more in this unit (10.2%) because of increased handling of the larvae and (d) more labour was involved for preparation of the second set of diet, transfer of growing larvae from one diet bottle to the second diet bottle and for the collection of pupae from individual vials (Table 1).

### Rearing in louvers

In this rearing unit, though pupal weight ranged from 315 to 390 mg (which was on par with individual rearing in diet bottles), the major disadvantages were: (a) only 20 to 30% survival was obtained, (b) insufficient aeration and fungus formation in the cells, (c) mortality due to cannibalism caused by the larvae moving from one compartment to the other and escape of larvae as the larvae had to be transferred into the open louvers and also as the covering sheet was not exactly fitting over the louvers (35.1%), (d) each compartment in the louver could hold only 5 gm of diet which was not adequate for complete larval development and so the larvae had to be transferred into another set of louver with diet; hence handling of larvae was more and disease incidence was high (30.3%) and (f) there was wastage of diet as in the case of rearing in diet bottles.

### Rearing in multicellular tray

The major advantages of using this tray were: (a) it could hold 100 larvae and a survival of 80 to 90% was recorded (b) the trays were made of readily available materials, (c) they were amenable to surface sterilisation and both the trays and lid could be reused, (d) each compartment could hold 11 gm of diet which was adequate for the total larval development and also there was enough moving space for the larva in each compartment, (e) by transferring 4 to 5-day-old larvae and by finally collecting the pupae, handling and disease incidence were minimum (1.5%), (f) the inverted top tray consisted of the compartments, because of which the larva in each compartment was totally separated from those in the neighbouring compartments; so the larvae could not migrate from one cell to another and also escape of larvae and mortality due to cannibalism were minimum (1.2%), (g) the mesh lined lid provided enough aeration and hence problem of moisture accumulation was not encountered, (h) the trays were transparent and thus pupal formation could be observed without disturbing the set up, by just inverting the tray (i) pupal collection was extremely easy - when pupae were formed, the top tray was removed, along with which major portion of the diet was also removed; majority of the pupae were then collected from the floor of the bottom tray and the few remaining pupae which were in the top tray could be separated out from the diet, (j) the trays could be stacked, (k) rearing in these trays involved minimum time and labour (Table 1) and (l) pupae were healthy and pupal weight ranged from 301 to 385 mg which was on par with that recorded in the individual rearing system.

The data provided on Table 1 show that the louver method was least suitable as the per cent pupation was extremely low. Besides, escape, disease incidence, involvement of time and labour and cost of rearing were high (Table 1). The performance of multicellular tray was on par with that of rearing in diet bottles with reference to per cent pupation, pupal weight, per cent adult emergence and prevention of larval escape. However, the cost of rearing 100 pupae in the multicellular unit was Rs. 118 in comparison to Rs. 156 in individual bottles. There was about 50% reduction in the amount of time spent in rearing when the multicellular tray was used. Though the cost of each multicellular larval rearing unit was high, because of the life of the tray, its reusability and efficiency, the rearing cost reduced considerably (Table 1). Therefore, among the three types of larval rearing units, the multicellular unit proved to be the most suitable.

Though individual rearing is widely used in research for production of high quality insects (Griffith and Smith, 1977; Teakle and Jensen, 1985), it is not economical for

large scale production. It is generally emphasised that for the development of any IPM package, it is essential that the insect rearing component has to be as 'local' as possible in design (Ocheing Odero *et al.*, 1991) and the multicellular larval rearing unit described above was also fabricated using indigenous materials. Bell (1991), Hartley *et al.* (1982) and Singh and Rembold (1992) found that multicellular larval rearing units were more advantageous for rearing *Heliothis* spp. in comparison to individual rearing in cups or bottles. In the rearing unit developed by Singh and Rembold (1992), the opening in each cell had to be covered using cotton wool, whereas the mesh lined lid in the present unit is a modification which has led to saving of time and labour.

It has thus emerged that the newly developed multicellular rearing unit is extremely useful for the rearing of *H. armigera* and could also be used for rearing other lepidopteran larvae like *Spodoptera litura* (Fabricius). It is suitable for production units where more sophisticated facilities are not available.

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## Influence of Prey Species on the Development and Reproduction of *Acanthaspis siva* Distant (Heteroptera: Reduviidae)

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**Abstract:** The prey insects such as *Spodoptera litura* Fab., *Earias vittella* Fab. and *Corypha cephalonica* Stainton were observed to exert a direct influence on the development and reproduction of the reduviid predator *Acanthaspis siva*. Maximum fecundity, longevity and minimal developmental periods were observed in *A. siva* when fed on the larvae of *S. litura*. Variation in the life table parameters of the predator was also observed on the three different prey species. The intrinsic rates of increase were 0.030, 0.028 and 0.024 per female per day for *S. litura*, *E. vittella* and *C. cephalonica* respectively. The population multiplied 33.78, 29.48 and 20.62 times in the cohort generation time of 155.383, 158.103 and 163.279 days for *S. litura*, *E. vittella* and *C. cephalonica* respectively.

**Keywords:** *Acanthaspis siva*, life tables, intrinsic rate of increase, *Spodoptera litura*, *Earias vittella*, *Corypha cephalonica*.

### INTRODUCTION

Augmentation of natural enemies, their mass rearing and release at appropriate stage and condition are major components in Integrated Pest Management (IPM). Successful culturing of suitable prey in the laboratory for rearing predators and parasitoids depends on the biology, behaviour and reproductive fitness of natural enemies. In order to augment this approach in biological control, increased attention needs to be diverted towards factors involved in successful predation, prey suitability as well as survivorship. Duration of the post embryonic development, fecundity, longevity and number of prey consumed during the life time are of paramount importance in assessing the predatory efficiency of an individual (Ananthakrishnan, 1996). The quality and quantity of nutrients of the prey influence not only the growth rate and survival of the predator (Ambrose and Subbarasu, 1988; Ambrose *et al.*, 1990; O'Neil and Wiedenmann, 1990; Ambrose and Rani, 1991) but also the fecundity and life table characteristics such as generation time as well as intrinsic rate of population increase (Awadallah *et al.*, 1986).

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One of the objectives of pest management is the estimation of the rate of growth of pests and their natural enemies (Howe, 1953) and life table studies is among the most useful numerical aid in studying population biology (Southwood, 1978) thereby enabling determination of age distribution and mortality rate in natural populations. *Acanthaspis siva* Distant has been recorded as an efficient predator on various economical important insect pests (Subbiah and Mahadevan, 1957; Ambrose, 1986; Ambrose and Livingstone, 1987; Lakkundi, 1989; Ambrose *et al.*, 1994). The prey insects significantly influence vital phenomena such as rate of development, survival and reproductive potential of insects which ultimately determine the rate of population build-up. Hence, life table studies of this predator on different insect pests are of significance in assessing the innate capacity for increase and information on population dynamics enables augmentation and subsequent release in the field for control of insect pests. The present investigation reports the life table analysis of *A. siva* on three common insect pests *Spodoptera litura*, *Earias vittella* and *Corcyra cephalonica*.

#### MATERIAL AND METHODS

Adults of *A. siva* were collected from Kumbakkai scrub jungle (foot hills of Ko-daikanal), Madurai district of Tamil Nadu, South India and reared in the laboratory on larvae of *S. litura*, *E. vittella* and *C. cephalonica* separately in 200 cc plastic containers. A cohort consisting of 100 eggs from each set was used to construct life tables. Eggs were collected and allowed to hatch in small plastic containers (60 cc) with moistened cotton swabs for maintaining optimum humidity (85%). The cotton swabs were changed periodically to prevent fungal attack. After hatching, all the nymphs were reared individually in plastic containers (60 cc) and fourth instar larvae of *S. litura*, *E. vittella* and *C. cephalonica* were provided as prey for respective cohort. Observations were made on hatching, completion of nymphal development, successful adult emergence, fecundity and age specific mortality in respective stages. Life tables were constructed according to the methods of Birch (1948) elaborated by Howe (1953), Watson (1964) and Southwood (1978).

In life table statistics, the intrinsic rate of increase was determined by using the equation  $\Sigma e^{-rm} X l_{mx} = 1$ , where  $e$  is the base of the natural logarithms,  $X$  is the age of the individuals in days,  $l_x$  is the number of individuals alive at age  $X$  as the proportion of 1, and  $mx$  is the number of female offsprings produced per female in the age interval  $X$ . The sum of the products  $l_{mx}$  is the net reproductive rate ( $R_0$ ). The rate of multiplication of population for each generation was measured in terms of females produced per generation. The precise value of cohort generation was calculated as follows:

$$T_c = \frac{\sum l_{mx} X}{R_0}$$

The arbitrary value of innate capacity for increase  $rc$  was calculated from the equation

$$rc = \frac{\log e R_0}{T_c}$$

This is an appropriate  $rm$  value. The values of the negative exponent of  $e^{-rm} X$  ascertained from this experiment often lay outside the range. For this reason both sides

Table 1: Biological data of *A. siva* on three insect pests ( $n = 20$ ;  $\bar{X} \pm SD$ )

Parameters (in days)	Prey Species		
	<i>S. litura</i>	<i>E. vittella</i>	<i>C. cephalonica</i>
Incubation period	18.600 $\pm$ 0.754	19.050 $\pm$ 0.826	18.100 $\pm$ 0.788
Nymphal duration	12.100 $\pm$ 0.640	19.100 $\pm$ 0.718	23.000 $\pm$ 0.858
I instar			
II instar	14.600 $\pm$ 0.598	17.250 $\pm$ 1.832	15.600 $\pm$ 0.598
III instar	14.950 $\pm$ 0.394	16.100 $\pm$ 1.334	18.111 $\pm$ 0.471
IV instar	21.053 $\pm$ 1.177	21.600 $\pm$ 0.707	23.125 $\pm$ 0.957
V instar	28.882 $\pm$ 0.928	32.704 $\pm$ 0.611	34.125 $\pm$ 2.125
Total developmental period	116.706 $\pm$ 3.274	126.438 $\pm$ 2.943	132.000 $\pm$ 2.386
Adult longevity	66.471 $\pm$ 13.408	59.125 $\pm$ 13.739	56.929 $\pm$ 13.539
Preoviposition period	20.134 $\pm$ 1.206	27.347 $\pm$ 1.879	29.843 $\pm$ 3.134
Fecundity*	114.125 $\pm$ 20.552	98.111 $\pm$ 17.892	83.875 $\pm$ 14.096

\*Number of eggs laid by a female in its lifespan

of the equation were multiplied by a factor of  $\Sigma_e^{7-rm} X l x m x = 1096.6$  (Birch, 1948; Watson, 1964). The two values of  $\Sigma_e^{7-rm} X l x m x$  were then plotted on the horizontal axis against their respective arbitrary  $rm$  on the vertical axis. Two points were then joined to give a line which was intersected by a vertical line drawn from the desired value of  $e^{7-rm} X l x m x$  (1096.6).

The point of intersection gives the value of  $rm$  accurate to three decimal places. The precise generation time (T) was then calculated from the equation.

$$T = \frac{\log eR_0}{rm}$$

The finite rate of increase ( $\lambda$ ) was calculated as  $e^{rm}$ . The weekly multiplication of predator population was calculated as  $(e^{rm})^7$ . The doubling time was calculated as  $\log 2 / \log \lambda$ .

## RESULTS

The incubation period of *A. siva* on *S. litura* was  $18.600 \pm 0.754$  days and it was extended to  $19.050 \pm 0.826$  and  $18.100 \pm 0.788$  days on *E. vittella* and *C. cephalonica* respectively. The total developmental period on *S. litura* was  $116.706 \pm 3.274$  days and it was extended to  $126.438 \pm 2.943$  and  $132.000 \pm 2.386$  days respectively on *E. vittella* and *C. cephalonica*. Increased adult longevity was evident in *A. siva* reared on *S. litura* ( $66.471 \pm 13.408$  days) than those individuals reared on *E. vittella* ( $59.125 \pm 13.739$  days) and *C. cephalonica* ( $56.929 \pm 13.539$  days). Maximum number of eggs per female was observed in *A. siva* reared on *S. litura* ( $114.125 \pm 20.552$ ). The egg number decreased to  $98.111 \pm 17.892$  and  $83.875 \pm 14.096$  when reared on *E. vittella* and *C. cephalonica* respectively (Table 1).

The life table parameters of *A. siva* on the three prey insects is given in table 2. The survival of adult female and the number of female births are shown in the Figures 1–3 which indicate that both the survival and the female birth of *A. siva* were found to

Table 2: Life table statistics of *A. siva* reared on different prey species

Parameters	Prey species		
	<i>S. litura</i>	<i>E. vittella</i>	<i>C. cephalonica</i>
Net reproductive rate ( $R_0 = \sum 1 \lambda_{mx}$ )	33.780	29.480	20.620
Mean length of generation ( $T_c = \sum t_{mx} X / R_0$ )	152.383	158.103	163.279
Innate capacity for increase in numbers ( $r_c = \log e R_0 / T_c$ )	0.023	0.021	0.019
Corrected rm ( $e^{T - rm X / \lambda_{mx}} = 1096.6$ )	0.030	0.028	0.024
(female/female/day)			
Corrected generation time ( $T = \log e R_0 / rm$ )	117.328	120.847	126.094
Finite rate of increase in numbers ( $\lambda = \text{anti log } e^{r_m}$ )	1.030	1.028	1.024
Weekly multiplication ( $e^{r_m 7}$ )	1.234	1.217	1.183
Doubling time ( $\log 2 / \log \lambda$ )	23.450	25.110	29.450

differ when reared on three different prey species. The highest survival and female birth were noted on *A. siva* reared on *S. litura* and the lowest on those reared on *C. cephalonica*. The net reproductive rate ( $R_0$ ) of *A. siva* was (33.98) significantly higher on *S. litura* than on *E. vittella* (29.48) and *C. cephalonica* (20.62). The intrinsic rate of population increase (rm) on *S. litura* was 0.030, decreasing to 0.028 and 0.024 on *E. vittella* and *C. cephalonica* respectively. The mean length of generation was shorter on *S. litura* (152.383 days) followed by *E. vittella* (158.103 days) and *C. cephalonica* (163.279 days). Consequent to the decrease in rm and extension of developmental period, the population doubling time on *E. vittella* and *C. cephalonica* increased to 25.110 and 29.450 days from 23.450 days of that reared on *S. litura*. The weekly multiplication rate on *S. litura*, *E. vittella* and *C. cephalonica* were 1.234, 1.217 and 1.183 times respectively.

## DISCUSSION

Differences in the value of biocontrol agents in the field can be predicted with caution, from measurements of biological parameters used in quality monitoring. Variation in the quantity of nutrients of prey species appear to have considerable effect on the feeding efficiency and reproductive potential of the predators (Beddington, 1975). The shortest developmental period of the predator *A. siva* was recorded when reared on *S. litura* which might be due to the minimum stress developed during predation on less number of preys due to the comparatively larger size with richer body tissue. Anderson (1962) reported a high variation in the rates of development and reproduction of six *Anthocoris* spp. when fed with a range of prey species. Awadallah *et al.* (1986) reported a high variability in the life cycle of *Xylocoris flavipes* when reared on five different prey species. Eggs, I and II instar larvae of beet armyworm proved to be inadequate food for *Podisus maculiventris*, while the fourth instar provided adequate nutrients for completing the life cycle of the predator (De-Clercq and Degheele, 1994).

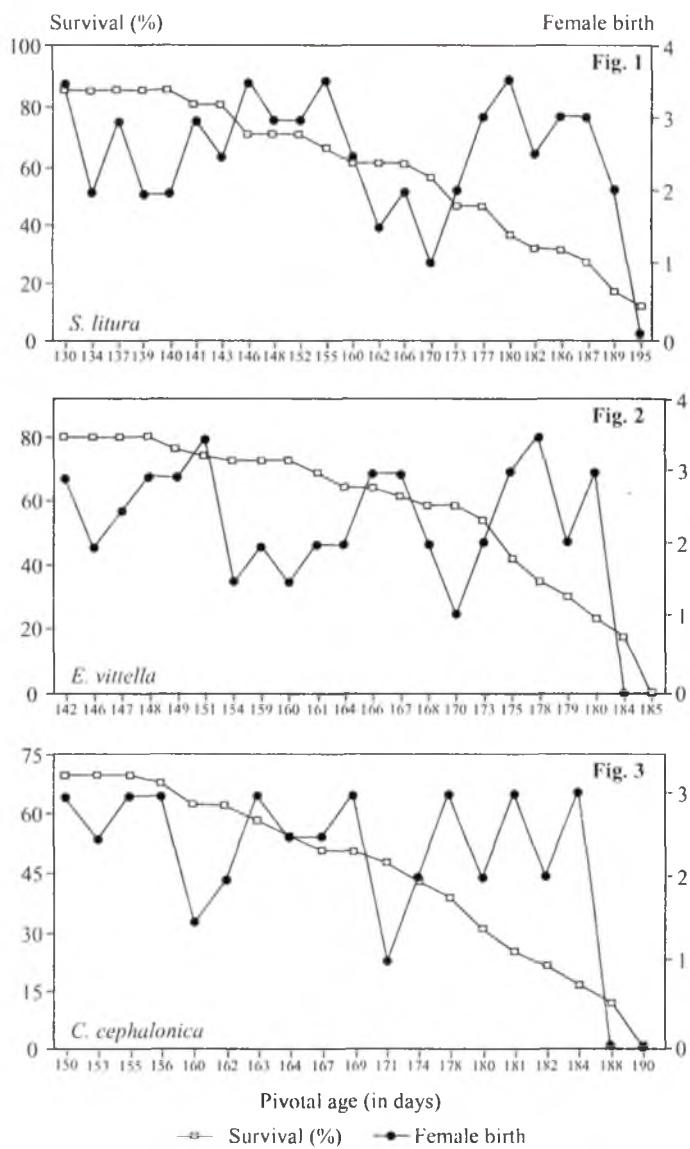


Fig. 1-3. Survivorship (1x) and natality (mx) patterns of *A. siva* cohorts at different prey regimens: 1: *S. litura* 2: *E. vittella* 3: *C. cephalonica*

Venkatesan *et al.* (1997) reported higher and faster development of reduviid *Cydnocoris gilvus* occurred on the prey *S. litura* than on *Oxya nitidula* and *Odontotermes obesus*. Increasingly, preferred prey accelerates food consumption, conservation and rate of development of the insects (Slansky, 1982).

Prereproductive delay and reproductive potential such as fecundity, percent hatch-

bility of insect predators are determined by the nutrient composition of the prey species (Fuller, 1988). Egg laying potential and longevity of adults were maximum on *S. litura* than on other prey species perhaps due to the higher primary nutrients. Reduced level of preoviposition delay, oviposition period and fecundity of *Menochilus sexmaculatus* were observed on the eggs of ants than on its natural host *Aphis craccivora* (Agarwala and Choudhury, 1995). A wider variation in the fecundity of *M. sexmaculatus* was reported owing to feeding on the adults of *Myzus persicae* and *Schizaphis graminum* (Haque and Islam, 1982). Extended longevity and higher fecundity were noted in *C. gilvus* on *S. litura* (Venkatesan *et al.*, 1997).

All life table statistics varied with the prey species. Natality and survivorship curves reflected the higher net reproductive rate ( $R_0$ ) of *A. siva* on *S. litura* than on other prey species. The  $R_0$  of the anthocorid *Lyctocoris campestris* varied with maximum values on pyralids and minimum values on *Trichoplusia ni* (Arbogast, 1983). Similarly, Awadallah *et al.* (1986) observed that the survivorship and fecundity of *Xylocoris flavipes* was maximum on *Tribolium castaneum* compared to *Oryzaephilus surina* as prey species. Higher survivorship and fecundity and less mean length of generation were noted in the reduviid *C. gilvus* on *S. litura* than on *O. nitidula* and *O. obesus* (Venkatesan *et al.*, 1997).

In the present study, *S. litura* was found to be the highly preferred prey of *A. siva* for the following reasons; a) faster development, b) higher survival, c) higher fecundity, d) higher net reproductive rate and e) shorter population doubling time of the predator. A perusal of literature reveals paucity of information on life table of predatory bugs. High net reproductive rate and intrinsic rate of population increase have been reported for temporate bugs such as *Oncopeltus fasciatus* (Klausner *et al.*, 1980), *Piegodorus guildnii* (Panizzi and Slansky, 1985) and *Clavigrelle tomentosicollis* (Iheagwam, 1982) when reared on their preferred hosts. Hence, considerable attention is needed to select the appropriate prey species for augmenting the insect predators in pest management practice.

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## Record of the Natural Enemies of the Lace Bug *Stephanitis typica* (Distant) a Pest on Coconut Palm.

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**Abstract:** Search for natural enemies of the lace bug *Stephanitis typica* revealed the presence of 14 insects and 23 spiders as predators and one fungal pathogen, in addition to the mirid predator *Stethoconus praefectus* recorded earlier. Two reduviids and one chrysopid were studied for their biology and feeding potential. *Endochus inornatus*, *Rhinocoris fuscipes*, *Ankylopteryx octopunctata octopunctata*, *Chelisoches morio*, *Phidippus* sp., *Tetragnatha andamanensis* and *Aspergillus flavus* are new records of natural enemies on *S. typica*.

**Keywords:** Lace bug, *Stephanitis typica*, natural enemies, predators, coconut

*Stephanitis typica* (Heteroptera : Tingidae) is one of the sucking pests on coconut palm foliage. It plays a vital role in coconut cultivation as it is one of the proved vectors of root (wilt) disease of coconut palm in India, a malady caused by phytoplasma (Mathen *et al.*, 1990). The immature and adult stages of the bug live on the abaxial surface of the leaves in colonies and drain the contents of the leaf tissue which leaves permanent dechlorophyllled yellow marks on the upper surface. Spraying with insecticides such as, 0.05% carbaryl or 0.01% endosulfan or monocrotophos (Nair *et al.*, 1991) and 0.03% phosphamidon (Borad *et al.*, 1993) checked the population of the lace bugs in the field. Many natural enemies were found associated with the lace bug in the field and recently attention was directed to the utilization of these natural enemies for the biological suppression of this pest. Among the natural enemies recorded, the mirid *Stethoconus praefectus* (Distant) was described and its biology studied (Mathen *et al.*, 1967; Mathen and Kurian, 1972). Livingston and Yacoob (1986) recorded a mymarid parasite *Parallelaptera* sp. on the eggs of *S. typica*. Balsdon *et al.* (1996) recorded another mymarid *Anagrus takeyanus* Gordh et Dunbar parasitising the eggs of *S. pyroides* (Scott), a pest of Azalea. Studies were taken up at this Institute to identify more bioagents and establish their role in controlling the lace bug. The present note summarizes the observations on the natural enemy fauna associated with *S. typica*.

Three locations viz. Ayiromthengu, Krishnapuram and Thottappally in Kerala were selected for the study. Collections were made every month, at random, from each

Table 1: List of natural enemies associated with *Stephanitis typica* on coconut palm

Order	Family	Fauna collected	Number of species collected	Location			Number of	
				I	II	III	Predator	Non-predator
Heteroptera	Miridae	<i>Stethoconus praefectus</i> (D)	1	+	+	+	1	0
	Reduviidae	<i>Endochus inornatus</i> * (Stal.)	1	+	-	+	1	0
		<i>Rhinocoris fuscipes</i> * Fabr.	1	-	+	-	1	0
		Unidentified	4	+	+	+	4	0
Neuroptera	Chrysopidae	<i>Ankylopteryx octopunctata</i> <i>octopunctata</i> * (Fabr.)	1	-	+	-	1	0
Coleoptera	Carabidae	<i>Parena nigrolineata</i> (Ch.)	1	+	-	+	0	1
		Unidentified	14	+	+	+	3	11
Dermaptera	Chelisochidae	<i>Chelisoches morio</i> * (Fabr.)	1	-	+	-	1	0
Orthoptera	Mantidae	Unidentified	1	-	+	-	1	0
Odonata		Do	2	-	-	+	0	2
Hymenoptera	Formicidae	Do	1	+	-	-	1	0
		Do	3	+	-	+	0	3
Diptera		Do	2	+	-	+	0	2
Araneae	Clubionidae	<i>Cheiracanthium</i> sp.	1	+	-	+	0	1
	Salticidae	<i>Phidippus</i> sp.*	1	-	+	-	1	0
	Tetragnathidae	<i>Tetragnatha andamanensis</i> *	1	+	-	+	1	0
		Tikader						
		Unidentified	23	+	+	+	21	2
Moniliales	Aspergillaceae	<i>Aspergillus flavus</i> Link*	1				pathogenic	

I=Ayiromthengu + = present \* = new records II= Krishnapuram - = absent III= Thottappally 0=nil

of these centres and all the fauna associated with lace bug colony/lace bug infested leaves of coconut palms were isolated and identified. The dead and diseased lace bugs were also screened for the occurrence of the pathogens. The insect and spider fauna isolated were further tested for their predacious/parasitic habits. The predators were then tested for their predator potential, biology and seasonal occurrence. The microorganisms isolated were also tested for their pathogenicity and the pathogenic forms were cultured for more detailed investigations.

During this study 33 insects and 26 spiders were observed (Table 1). Among these 14 insects and 23 spider species were observed as predators on lace bug. In addition, *Aspergillus flavus* Link was found to be pathogenic to the lace bug nymphs and adults. These natural enemies except *S. praefectus* are new records on *S. typica*. Of the fourteen predacious insects collected, *Endochus inornatus* (Stal.), *Rhinocoris fuscipes* Fabr. and *Ankylopteryx octopunctata* (Fabr.) were studied for their biology and feeding potential.

#### *Endochus inornatus* (Stal.)

Adult bugs are brown in colour. *E. inornatus* completes its egg period in 7–10 days and nymphal period 41–81 days in five instars in the laboratory, at a temperature and humidity range of 26–33°C and 77 to 93%, respectively. Females started laying eggs in 16–23 days after emergence and the fecundity ranges from 230–355 eggs per female. Eggs are laid in groups of 10–38 per batch and only the first laid eggs are cemented

to the leaves. Rest of the eggs are glued to the first laid eggs to form a group. Eggs are cylindrical and brown in colour with white operculum bearing a hair-like structure. The early instar nymphs consumed 5.9–7.9 lace bugs per day. The predator nymphs and adults also feeds on the larvae of the rice moth *Corcyra cephalonica* (Stainton) in the laboratory. *E. inornatus* is observed in the field from April to September.

#### ***Rhinocoris fuscipes* Fabr.**

Adult bugs are orange-red in colour. Eggs are brown in colour with white operculum. Eggs are laid in batches consisting of about 38 and are seen cemented to the leaves. Nymphs are orange-red in colour with yellowish-green legs bearing brown spots. Nymphal period is completed in five instars in 26–40 days. Predator nymph consumed 5–7 and 15–29 lace bug nymphs during I and II instar, respectively. They also consumed larvae of *C. cephalonica* in the laboratory.

#### ***Ankylopteryx octopunctata octopunctata* (Fabr.)**

*A. octopunctata octopunctata* is predacious on all stages of lace bug. It occurs in the field during all months of the year with a peak in April. The females lay pedunculate eggs in batches of 20–25 on coconut leaves. The predator completes its egg to adult period in 22–27 days which comprises of an egg period of 3–5 days, larval period of 14–18 days and pupal period of 5–7 days. The larva of the predator feeds on the nymph and adult lace bugs, their consumption varied from 6–39 lace bug/predator/day. It also consumed *Corcyra* eggs @ 11–185 per day. Adult chrysopid feeds on honey.

Reduviids and chrysopids are known predators on a number of crop pests. *E. inornatus* is recorded as predator on *Helicoverpa armigera* (Hubner), *Helopeltis antonii* Signoret and *Idioscopus* spp. (CPCRI, 1982; Singh, 1994; Singh *et al.*, 1993). Another species of *Endochus* is recorded as a predator on larval stages of *Hyblaea puera* Cramer (Mohanadas, 1996). *R. fuscipes* is a polyphagous predator. It was recorded as a predator on the ash weevil *Myllocerus curvicornis* (F.) on coconut (Ponnamma *et al.*, 1979). Similarly it was also recorded as a predator on *Diacrisia obliqua* Walker (Singh and Gangrade, 1974); on *Nezara viridula* Linn. (Singh and Rawat, 1982); on *Dicladispa armigera* (Olives) (Singh, 1985); on pests of cotton and maize (Ambrose and Livingston, 1986) and on *H. armigera* (Vennison and Ambrose, 1986). Sathiamma *et al.* (1985) recorded *A. octopunctata candida* as a predator on the eggs and larvae of *Opisina arenosella* on coconut palm. The mirid predator *S. praefectus* occurs during all months of the year with peak during March and August. This predator completes its egg to adult period in 15–17 days and prey consumption ranges from 3–30 lace bugs/predator. It is one of the promising natural enemies of *S. typica* in the field (Mathen and Kurian, 1972). Combined with the reduviid, chrysopid and spider predators, these mirids are capable of suppressing the population of the lace bugs considerably in the field.

#### **ACKNOWLEDGEMENTS**

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## **Biology and Breeding Behaviour of the Elephant Dung Beetle, *Helicocoris dominus* Bates (Coprinae: Scarabaeidae)**

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**Abstract:** The nest of *Helicocoris dominus* Bates consists of a tunnel going down into the soil and opening into a horizontal, ovoid brood chamber in which each brood develops inside individual brood balls (usually 3), constructed by the female out of fresh elephant dung. The brood chamber is generally isolated from the main mortality factors and fluctuations of outside temperature and humidity. The mother beetle continues to live inside the brood chamber, taking care of the developing larvae until pupation, contributing to the reproductive success of this subsocial insect.

**Keywords:** Biology, breeding behaviour, nidification, control of clutch size, *Helicocoris dominus*

*Helicocoris dominus* Bates is the largest of the 3 species of this genus occurring in India. This species depends exclusively on the fresh dung of wild elephants for feeding and breeding. Its distribution therefore coincides with the habitats of the wild elephants. The field work for the present study was carried out from June 1989 to April 1991 in the elephant habitats of the Karulai Forests of the Nilambur Subdivision (Latitude: 10°09'–11°26' N and Longitude: 75°48'–76°33' E), Kerala. During their emergence period (early June to end of July) these beetles were also collected at light on fresh elephant dung. The pronounced sexual dimorphism and intra-sex variations of this species were studied by Joseph (1994).

**Emergence of adults.** The development of *H. dominus* from egg to adult takes place inside a brood ball, lodged in a brood chamber, located in the soil at a depth varying from 27–56 cm (Fig. 1). The adults of the new generation start emerging from the soil usually from the first week of June, 2–3 days after heavy rains of the South-West monsoon, when the brood balls in which they have developed, as well as the soil above, are adequately water-soaked. Beetle emergence takes place after nightfall, normally from 18 to 21 hours. Emerged beetles soon fly to fresh elephant dung pats to which they are known to be very strongly attracted by the dung odour (Shibuya and Inouchi, 1982).

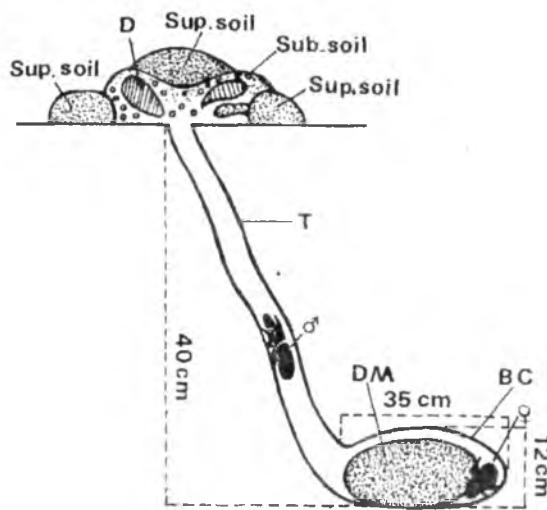


Fig. 1: Vertical Section (diagrammatic) through dung pat and completed tunnel and brood chamber of *H. dominus*. D: dung; Sup. soil : superficial soil; T: tunnel; B.C: brood chamber; D.M: dung mass.

*Maturation feeding.* Emerged adults of both sexes (Fig. 2) attain maturity for breeding after a period of 'maturation feeding' (or 'Reifungsfrass' - period) lasting approximately 4 weeks inside separate feeding tunnels excavated below fresh dung pats and packed with dung.

#### Breeding behaviour and nidification.

*a) Excavation of brood burrow (nest).* Push ups of soil, lying adjacent to fresh dung pats (Fig. 3), indicate that the beetles are engaged in excavation work for nidification. Often up to 3 beetles have been seen digging their separate tunnels below a dung pat, and all such individuals are females. The fully excavated tunnels of *H. dominus* are elliptical in outline (around 4.5 cm  $\times$  4 cm) and descend at an angle of about 45° below the surface of the soil to a depth of 27 to 56 cm ( $42.53 \pm 7.32$ ;  $n = 20$ ). At this level each tunnel opens into a large, ovoid brood chamber with length varying from 19 to 50 cm ( $31.75 \pm 8.02$ ), width from 9.5 to 19.00 cm ( $14.13 \pm 2.32$ ) and height from 10.00 to 18.00 cm ( $13.64 \pm 1.99$ );  $n = 20$ . (Fig. 1). Observations on the sequence of events taking place inside the brood chambers were made possible by excavating marked brood burrows (nests) of several of these beetles in different stages of their nesting, provisioning and brood ball construction.

*b) Provisioning.* After completing the tunnel and the brood chamber, the female transports dung from the overlying dung pat into her brood chamber in the manner as found by Kingston and Coe (1977) in the african elephant dung beetle *H. dilloni*. Each mass of dung that is pushed down is compacted with force against the distal end of the brood chamber. This process is repeated until sufficient dung to make 3 or 4

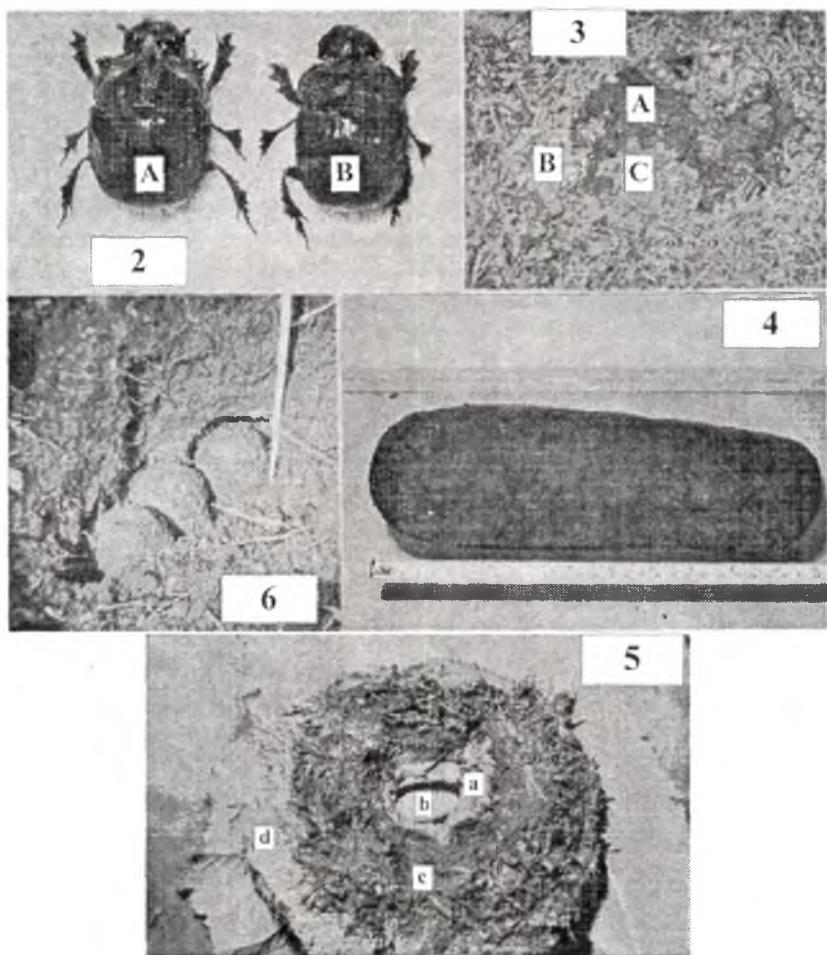


Fig. 2: Adults of *H. dominus*. A: Male; B: Female; 3: Push-ups of soil adjacent to dung pat indicating nest excavation below. A: dung; B: superficial soil; C: sub-soil.; 4: The compacted, smooth, round-ended, cylindrical dung mass taken out from a brood chamber.; 5: Vertical Section of a finished brood ball. a: egg-pouch lined with a layer of soil; b: egg *in situ*; c: provision of dung; d: outer coating of soil.; 6: An overview of the brood chamber showing 3 brood balls *in situ*. The steel tape (in white) indicates the depth of the chamber in the soil.

brood balls which are stored in the chamber in the form of a single, compact, smooth, round-ended cylinder (Fig. 4). The length of this dung mass varies from 15.50 to 39.00 cm ( $27.07 \pm 6.23$ ); width varying from 8.00 to 10.30 cm ( $9.23 \pm 0.67$ ); weight varying from 880 to 2473 g ( $1529.05 \pm 426.05$ );  $n = 20$ .

No male has been found anywhere in the nest to help the female during her arduous task of excavation and provisioning. But once provisioning is over, a male invariably appeared in each tunnel, most probably in response to the female's acoustic signals

(Joseph, 1991). Copulation has not been observed in *H. dominus* or in any other species of this genus. The female now excavates in the wall of the brood chamber in front, above and behind the dung mass, and in this process, a thin soil layer comes to adhere to the whole exterior of the dung mass which now lies free on the floor of the brood chamber, with a 4 to 5 cm wide air space in front, above and behind it.

*c) Brood ball construction.* In nests more than 15 days old, the females start brood ball construction. The cutting up of the compact dung mass into submasses (each of which eventually is incorporated into a brood ball containing a single egg) is carried out progressively (from the end nearest the entrance to the brood chamber), sequentially, and over a period of several days. Further details of brood ball construction behaviour and of the finished product (Fig. 5) are very much similar to the same in *H. dilloni* (Kingston and Coe, *op. cit.*).

Out of a total number of 51 brood chambers examined, 35 contained 3 brood balls in each (Fig. 6), 9 had only 2, and 7 had 4. The number of brood balls in a brood chamber has been found to vary from 2 to 4 ( $2.96 \pm 0.56$ ;  $n = 51$ ). Some of the brood balls are subspherical (with diameters varying from 9 to 13 cm), while others are somewhat pear-shaped (diameters from 10 to 13 cm; lengths from 12.5 to 15.5 cm). The weights of the brood balls taken from the 51 brood chambers vary from 530 to 1750 g ( $958.68 \pm 250.01$ ;  $n = 151$ ).

*d) Control of microclimate, mortality factors, clutch size.* The microclimatic conditions in the brood chambers of *H. dominus* could not be measured due to certain technical problems. Kingston and Coe (*op. cit.*) reported that in *H. dilloni* even in brood chambers that are located at depths around 25 cm in soil, the temperature and humidity remained fairly stable throughout the developmental period, at 28°C and 8% respectively.

Based on the classification of the breeding biology of dung beetles (*Scarabaeidae*) proposed by Halffter and Matthews (1966), it may be stated that all species of the genus *Helicocoris* are subsocial. As a result of the highly organised breeding behaviour and nidification including well provisioned brood ball construction in *H. dominus*, the brood develops in an environment generally isolated from the main mortality factors (like parasitoids, predators, paucity of food and fluctuations of microclimate). Furthermore, the mother beetle continues to live inside her brood chamber, taking care of the developing larvae inside their brood balls until they pupate. This active maternal care (Brütpflege) serves significantly to boost brood survival, leading to reproductive success.

The available records within the *Scarabaeidae* suggest that there is a close adaptive relationship between the mode of breeding behaviour and nidification on the one hand and the number of eggs laid (clutch size) on the other, i.e., the more highly organised the breeding behaviour and nidification, the lesser the clutch size, and vice-versa. It is therefore reasonable to suggest that *H. dominus*, having a mean clutch size of 3 eggs, has a more highly organised breeding behaviour and nidification than *H. dilloni* that has a mean clutch size of 5 eggs. According to Klemperer (1983), the adaptive control of clutch size in the *Scarabaeidae* is induced by the brood, acting through the inhibitory action of brood pheromones inhibiting oviposition and follicle formation, and also by releasing parental care of the brood.

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## Survival Capacity of *Zygogramma bicolorata* in Diapause Condition in relation to Delayed Monsoon Showers

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**Abstract:** *Zygogramma bicolorata* adults undergo diapause in the soil from October to May, emerging with the onset of monsoon rains. A survey was carried out to determine whether delay in rainfall could affect emergence of diapause adults. It was found that delay of rainfall for more than 45 days could reduce the emergence of adults significantly. In such areas, fresh releases may have to be carried out for effective control of the weed.

**Keywords:** *Z. bicolorata*, diapause, delayed monsoon

Field releases of *Zygogramma bicolorata* Pallister (Coleoptera: Chrysomelidae) introduced from Mexico for biological control trials against *Parthenium hysterophorus*, have resulted in successful control of the weed in different parts of the country; whereby enhancing growth of the native flora (Jayanth and Geetha Bali, 1994; Jayanth and Ganga Visalakshy, 1995). In the field, the insects are active from June to October. Later the adults enter the soil and remain in diapause from November to May. With the onset of monsoon rains, they emerge, which also coincides with the appearance of the weed (Jayanth and Geetha Bali, 1993). Many a times, the monsoon rains get delayed in many parts of the country, where *Z. bicolorata* were released. A study was made under laboratory conditions to determine the survival and emergence of diapausing *Z. bicolorata* adults in relation to delayed rainfall.

*Zygogramma bicolorata* adults were collected during October – November from defoliated parthenium plants infesting I.I.H.R. farm and released into diapausing jars under laboratory conditions. The diapausing jars were plastic jars of 10 × 7 cms with wiremesh lid for aeration. The jar was filled upto 5 cm from the base with sterilised soil, which was moistened sufficiently, to enable the adults for burrowing. After 2–3 days the adults which have not entered the soil for diapause were removed. The diapausing jars were kept as such under laboratory conditions till the next monsoon shower.

Table 1: Survival and emergence of *Z. bicolorata* in relation to delayed rainfall.

No. of days in diapause	% emergence	No. dead in soil (%)
176	55.18 (47.9) <sup>a</sup>	12.75
192	45.84 (42.7) <sup>b</sup>	11.50
224	42.31 (40.4) <sup>b</sup>	17.70
252	4.23 (11.5) <sup>c</sup>	67.70
275	0.00 (0.00) <sup>c</sup>	53.00
281	0.00 (0.00) <sup>c</sup>	62.40

(The values given in parenthesis are angular transformed values).

Values followed by similar superscript are not statistically different at 0.05% level.

With the onset of monsoon rains during April – May, *Z. bicolorata* were observed in the field. Coinciding with this, some of the jars with diapausing adults were moistened enough, to facilitate adult emergence. Later the remaining jars were moistened initially at fortnightly and later at monthly intervals. Observations on the number of adults emerged was made, upto a week after moistening. Once emergence was stopped, the soil was examined for the remaining adults. The experiment was replicated thrice, with 100 adults per treatment. (Treatment were no. of days in diapause). The results were subjected to Anova test.

When jars with diapausing adults of *Z. bicolorata* were moistened, coinciding with the onset of monsoon, about 55.18% emergence was obtained (Table 1). Delay in rainfall by another 45 days did not affect the emergence, significantly. Thus, 45.84% and 42.31% emergence was obtained, at 192 and 224 days respectively. However, further delay caused high mortality. Thus after 252 days only 4.32% emergence was recorded. No adult emergence was obtained after 275 days. Examination of the soil revealed 12.75–62.40% dead adults for the above periods.

The above study reveals that delay of monsoon rains upto 45 days may not affect emergence of *Z. bicolorata* from diapause. However further delay could significantly reduce adult emergence. In such situations, fresh releases may have to be carried out to suppress the weed effectively.

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## Longevity and Age Specific Fecundity Of The Pea Aphid, *Aphis craccivora* Koch (Homoptera: Aphididae) On Cowpea

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**Abstract:** Longevity and age specific fecundity of *Aphis craccivora* was constructed. The intrinsic rate of increase (0.272) and true generation time (9.78 days) of the pest were found conducive to its population build up than those of *Lipaphis erysmi* reported earlier.

**Keywords:** Life table, *Aphis craccivora*

Pea aphid *Aphis craccivora* Koch is a very common pest of a variety of plants found throughout India. Lab-lab, ground nut, green gram, blackgram and many other pulses are often seen seriously damaged by this insect. The nymphs and adults infesting gregariously the tender shoots, inflorescence and tender pods cause malformation, stunting and even drying up of these parts.

For evolving a sound integrated control strategy against a pest, the basic information on the biology and ecology of the pest concerned is essential. The studies conducted on these aspects relating to *Aphis craccivora* in Kerala are quite limited. Hence studies were taken up to work out the longevity and age specific fecundity of the pest.

Life and fertility table of *Aphis craccivora* was prepared following the procedure of Phadke (1982).

Leaves of same age and size were selected from cowpea plants growing in pots. A day earlier, 2 mother aphids had been earmarked. Newly emerged nymphs from the earmarked aphids were transferred using a fine camel hair brush, to the selected leaves at the rate of four per leaf. Nymphs were then enclosed in microcages. Six replications were maintained. The survival of individual nymphs was observed daily at 4 pm till the natural death of all the individuals occurred. The number of young nymphs reproduced parthenogenetically by the emerging adults were also recorded every day. After counting, the newly emerged nymphs were carefully removed without disturbing the mother aphid and micro cages were again put back in position for further observations. All the newly born nymphs were found to be females only.

Table 1: Age specific fecundity of *A. craccivora* on cowpea

<i>X</i>	<i>Ix</i>	<i>mx</i>	<i>Ix.mx.</i>	<i>x.Ix.mx</i>
0	0	0	—	—
1	0.9583	0	—	—
2	0.8750	0	—	—
3	0.8750	0	—	—
4	0.7500	0	—	—
5	0.7500	0	—	—
6	0.708	2.66	1.880	11.28
7	0.708	0.66	0.467	3.26
8	0.666	2.00	1.320	10.56
9	0.666	1.66	1.090	9.81
10	0.580	1.83	1.060	10.60
11	0.458	1.50	0.687	7.55
12	0.416	10.00	4.160	49.92
13	0.416	2.00	0.832	10.81
14	0.416	3.80	1.580	22.12
15	0.333	2.66	0.877	13.15
16	0.333	1.00	0.330	5.28
17	0.166	0.16	0.025	0.43
18	0	0	0	0

Net reproductive rate ( $R_0$ ) = 14.308Approximate generation time ( $T_c$ ) = 10.82 daysCapacity for increase ( $rc$ ) = 0.245Intrinsic rate of increase ( $rm$ ) = 0.272Finite rate of increase ( $\lambda$ ) = 1.87True generation time ( $T$ ) = 9.78 days

Survival value ( $Ix$ ) and fecundity rate ( $mx$ ) at each pivotal age ( $x$ ) were worked out. The net reproductive rate ( $R_0$ ) =  $\sum Ixmx$ ; approximate generation time ( $T_c$ ) =  $\sum xIxm / R_0$ ; capacity for increase ( $rc$ ) =  $\log_e R_0 / TC$  and the intrinsic rate of increase ( $rm$ ) was calculated using the method of Birch (1948). The intrinsic rate was determined by using the relationship  $\sum e^{7-rm \cdot x} \cdot Ixm = e^7 = 1097$ . From this the finite rate of increase  $\lambda$  = antilog  $e^{rm}$  and true generation time ( $T$ ) =  $\log_e R_0 / rc$  were also worked out.

Age-specific survivorship ( $Ix$ ) and age-specific fecundity ( $mx$ ) are presented in the table. There was 29.2 per cent mortality during the nymphal period of six days and hundred per cent mortality at the 18th day. Age-specific fecundity was maximum (10) on the 12th day followed by 3.8, 2.66, 2.66, 2 and 2 on the 14th, 15th, 6th, 8th and 13th day respectively. Reproduction continued upto the 17th day.

The net reproductive rate was 14.308 during an approximate generation time of 10.82 days and the true generation time was worked out to be 9.78 days. i.e. the pest multiplied 14.308 times during a generation time of 9.78 days.

The intrinsic rate of increase ( $rm$ ) was 0.272 and finite rate of increase ( $\lambda$ ) was 1.87 per female per day. The population increase of *A. craccivora* seems to be much faster than that of *Lipaphis erysimi* as reported by Phadke (1982). The life cycle of the most efficient predator *Chilomenus sexmaculata* was 48.7 days with an average fecundity of 15.27 (Reji Rani, 1995).

Short life cycle and high fecundity of the aphid in comparison to that of the predator might result in rapid build up of the pest than the predator in the field and hence the failure of the predators in exerting control effect on pest population.

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## New Record of a Pupal Parasitoid *Perilampus nesiotes* Crawford from a Tachinid Parasite of *Anthraea proylei* (Jolly) (Lepidoptera: Saturnidae) from Manipur.

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**Abstract:** *Perilampus nesiotes* Crawford, a pupal parasitoid on *Blepharipa zebina* (Walker), a tachinid fly infesting oak tasar silkworm, *Anthraea proylei* in Manipur is reported for the first time.

**Keywords:** *Perilampus nesiotes*, pupal parasitoid, *Blepharipa zebina*, *Anthraea proylei*

*Perilampus* species are primary or secondary parasites of a wide range of hosts including saw flies (Hymenoptera), Neuroptera, Lepidoptera or Orthoptera. This genus is cosmopolitan in distribution. So far, 11 *Perilampus* species have been reported from various parts of India (Subba Rao and Hayat, 1986).

During 1997 spring rearing season, an ectopupal parasitoid, *Perilampus nesiotes* Crawford (Hymenoptera: Perilampidae) was collected from the puparia of *Blepharipa zebina* (Walker) (Diptera: Tachinidae) which is a serious pest of oak tasar silkworm, *Anthraea proylei* (Jolly) in Manipur. This parasitoid is noted to be solitary in status. Crawford (1911) described this species *P. nesiotes* from Philippines and Sumatra.

### ACKNOWLEDGEMENT

The authors are thankful to Professor T. C. Narendran, Department of Zoology, University of Calicut, Kerala, India for identification of the parasitoid.

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## ON RETIREMENT OF THE FIRST AAE PRESIDENT .....

Prof. M. R. G. K. Nair is retiring from his position as the President of the Association for Advancement of Entomology (AAE) from 1999 after being steering the Association since its inception for more than the last 24 long years. Prof. Nair who endeared himself to all his students, colleagues and other as MRGK has a very successful and academically rewarding career on all these years. Born on 21st February, 1922 and after having his preliminary education in his home state Kerala had his Masters degree from the University of Lucknow, Doctoral degree and subsequent training in Crop Nematology from IARI, New Delhi. He started his professional career as a Research Officer in Entomology in the year 1946 in Kerala University. Later when the College of Agriculture was started at Vellayani in 1955, he was shifted to this Institute and there he became Professor of Entomology in 1963. Later he was appointed as the Director of Research in Kerala Agricultural University in 1972. After his retirement in 1977 he served as ICAR Emeritus Scientist for 2 years. Invited by World Bank and FAO Prof. Nair visited Sri Lanka organizing Workshops to acquaint Agricultural Officers with the philosophy and practice of Integrated Pest Management.



Prof. Nair has been a pioneer in the development of agriculture education and research in Kerala especially in the field of Crop Entomology and Pest Control. He provided the initiative and leadership in the organization of a full fledged Department of Entomology in the college of Agriculture, Vellayani with excellent facilities for research/teaching on related entomological aspects viz: Biology, Ecology, Toxicology, Pest Control, Insect Pathology, Biological Control and Storage Entomology. He has a wide range of publications of his credit. Authored over 90 scientific papers based on his research findings on Crop Entomology and 3 books including a Monograph on **Crop Pests of Kerala** which received special attention among agriculturists and entomological scientists alike. Also received an All India Award for his Malayalam book **Karshikakeedavijnanam** from the University Grants Commission. He was a member of the ICAR Scientific Panel, member of the first Executive Committee of KAU and Agricultural Expert in the Board of Directors of State Bank of Travancore. Prof. Nair has earned himself a prominent place in the field of Indian Agricultural Entomology. After serving AAE for the last 24 years as President and now as he is 77 years of age wants to retire from his active academic activities.

It is with high respect and regard that me as the Secretary-Treasurer of AAE and the Managing Editor of the Journal ENTOMON, acknowledge with gratitude and put on record the leadership rendered by him in the steady and leaping progress attained by AAE during all these years. AAE wishes Prof. MRGK all the best in the years to come.

D. Muraleedharan  
Managing Editor  
ENTOMON

ENTOMON gratefully acknowledges the following reviewers, who offered their valuable time and expertise in critically evaluating the manuscripts during 1997-98.

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# ELECTRONIC VERSION OF ENTOMON

## INTRODUCTION

In 1665 *Journal de Scavants*, the first scholarly journal, was launched followed a few months later by the *Philosophical Transactions of the Royal Society of London* which is still in existence. Since then, very little has changed in the scholarly publishing industry. However, in the last few years, changes abound, fuelled by the advent of World Wide Web. Publishers realize that on the one hand the market is demanding electronic publishing, and that on the other hand they can do a better job with more efficient production and distribution. What is the long-term outlook?

## WHAT ARE PUBLISHERS DOING NOW?

Often headed by the large learned societies, publishers have instigated many initiatives on the Web. These include

1. Altering services on the Web: these are mostly free of charge, and include lists of contents and often abstracts. Surrounding infrastructures such as search engines are also provided. Good examples can be found on the AGU web pages (<http://www.agu.org/pubs/inpress.html>). The National Center for Petroleum Geology and Geophysics offers an excellent overview of the web-presence of earth science journals (<http://www.ncpgg.adelaide.edu.au/journals.htm>). Publishers hope that giving this service to the potential readers will lead to an increased readership and exposure of their journals.
2. Publish "unprintable" items on the web, such movies, sound, or computer programmes which are connected to conventionally published articles. Elsevier publishes sound files with journals for instance, *Speech Communication*.
3. Making existing journals electronic. Springer (<http://link.springer.de/ol/gol>) has most of its journals in pdf format which basically offers readers the possibility of reading (or printing) the articles in exactly the same form as they are printed in their respective journals.
4. Real electronic journals in HTML format. One early example would be *Earth and Planetary Science Letters* (<http://accept.elsevier.nl/journals/eps1/Menu.html>), which features electronic datasets, and also provides the abstracts of the literature references.

## IS ALL THIS REALLY ANY BETTER?

The above services do have their nice aspects, but on the whole we do not necessarily believe that they will undermine the popularity of the printed journal issue. Yes, it is convenient not to have to go to the library anymore to read a journal, but the amount of full-text journals available from the desktop is still very low. And, as most www publications imitate the printed product, how can one improve on the magnificent interface offered by the print medium? This has developed from the ancient times of clay tablets and book scrolls and now the interface of a paper issue has not only page numbers, a list of contents and keyword indexes but also ease of reading, convenience of flicking through pages, and portability. There is simply no comparison with a

computer screen, which is not portable, awkwardly hard to read, even harder to browse, may have inexorably slow connection times, and from time to time a breakdown. Uncertainties also exist concerning the future: for example, will a pdf file still be eligible with the equipment that will be used in 2098?

## OUTLOOK

One could anticipate that for a successful electronic product there should be emphasis on completeness, and on the additional values of the features offered by web-technology, I believe that the following points are essential ingredients for this:

1. **Complete offering and easy usage:** Scientists are more likely to go to electronic literature, once this offers a fairly complete overview. Thus, publishers should link their services using the same standards for representation and interface.
2. **To achieve eternal preservation, articles should be stored in a generic format.** Many publishers (including the IEEE, ACM and Elsevier) adhere to SGML (Standard General Mark-up Language), which allows them to generate articles in a *contemporary* format (at this time is conventional print, pdf and HTML3.2; five years ago it was postscript and HTML1.0. With SGML the text is exhaustively coded (tagged). For instance, literature references are tagged, detailing author name, journal, publication date and so on. This allows a publisher to link references to abstracts automatically, and even to link to the referenced article.
3. **Good searching methods** — not a search engine based on inanimate-like boolean logic, but a “fuzzy logic” search mechanism, using natural language as well as good thesaurus. Who knows, may be finding the information will become more important than the information itself.
4. **Link the primary literature with secondary literature.** Wouldn't it be great to do searches on information servers, and be hyperlinked immediately with the full-text of articles resulting from your search? And conversely, while reading an article, being able to click on the literature references and go to the abstract (or better still, to the article in full text — if within the database).
5. **Use the medium to the fullest.**
  - \* Multimedia files, such as animations, movies and sound; Java code
  - \* “Living” articles; articles that can be continuously revised and updated after publication
  - \* Computer programme code, plus test runs.

A journal publication featuring a number of the above points is *Earth & Planetary Science Online*, or *Bulletin of Mathematical Biology* which boasts a link with a secondary database and has HTML and pdf articles generated SGML code, as well as multimedia files (called datasets).

In order to put our journal on the web we have to devise a system that needs to be agreed upon and production of impeccable SGML code needs to be arranged with the typesetter. Our model is that of Elsevier Science, which now has 100 life sciences journals available in HTML/pdf in the project “science direct” (<http://www.sciencedirect.com/>), which has a direct link with EMBASE, Elsevier's abstracting/indexing system in the life sciences. We hope to extend this in a limited way to our journal, and I hope Entomon (with links to the abstracting/indexing services will soon be included).

The traditional added values of the publishing industry, such as composing, printing and distributing of articles are vanishing. And, although the paper publication medium is hard to beat, I am sure that the publishing industry is developing electronic publication systems which



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